TROPICAL DISEASES BUREAU.

TROPICAL VETERINARY BULLETIN.

No. 2.] 1913. [Vol. 1.

ANAPLASMOSIS.

(114) BEVAN (Ll. E. W.). **Anaplasmosis of Sheep.**—*Veterinary Jl.* 1912. July. Vol. 68. No. 445. pp. 400-401.

Several outbreaks of disease occurred among sheep in March of this year, the most noticeable symptom in every case being a dropsical condition of the throat. In some cases this has been the only symptom mentioned, but in others the dropsy has been more severe and extensive.

In one outbreak investigated by the author parasites were found in the red blood corpuscles resembling the Anaplasma of cattle. In addition, all the changes usually associated with severe anaemia were observed in the blood, the number of cells having fallen to nearly half the normal.

It would appear that the disease has a wide distribution throughout Rhodesia.

(115) CARPANO (M.). **L'Anaplasmosi nei Bovini della Campagna Romana. (Nota Preventiva).** [Anaplasmosis in the Roman Campagna. Preliminary Note.]—*Il Moderno Zooiatro* (Parte Scientifica). 1912. Aug. 31. Vol. 23. No. 8. pp. 336-342. With 1 text-figure.

The case was observed by GABBUTI who suspected piroplasmosis, but since the train of symptoms usually observed in cases of *bigeminum* infection was not present blood examinations were made by the author. The animal's temperature at the time of the blood examination was 40·4° C.

GABBUTI gave the author details of six similar cases that he had observed in the same district the previous year. Of these cases five terminated fatally. The disease was met with principally in well-bred imported animals and especially in cows near to calving.

Clinical characters of the disease.—The onset of the disease is marked by a rise of temperature which persists with exacerbations during the whole course of the attack. The appetite is capricious, and rumination may be entirely suspended. The faeces are

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normal in consistence but contain a large quantity of mucus. The urine is normal in tint. The conjunctival, buccal, and vaginal mucous membranes are pale and anaemic, but no petechiae or extravasations have been observed.

In the fatal cases death occurred about the 12th to 15th day, and a few days before death there was haemoglobinuria.

In non-fatal cases the temperature falls within two or three days to normal with a few slight exacerbations.

Transmission of the disease.—Since specimens of the *Rhipicephalus bursa* were found upon the cow it appears to be possible that that is the transmitting tick.

Examination of the blood.—A blood count showed that there were about three million erythrocytes per cubic millimetre and about eight thousand leucocytes.

Microscopic examination was made of the blood in fresh and in stained preparations.

Fresh preparations.—In unstained fresh preparations the small size of the parasites made accurate examination difficult. They appeared as somewhat refractile points at the periphery of the red corpuscles. The diplococcus forms appeared to show a slow rotary movement.

Stained preparations.—The best results were obtained with specimens fixed in sublimate alcohol and subjected to prolonged staining with Giemsa (1 in 40), with subsequent decolourisation.

Anaplasma is ordinarily a parasite of the red corpuscles but it may be found free in the plasma. In the majority of cases the parasite is disposed towards the periphery of the host cell, but it may be observed in the centre.

In well differentiated specimens each parasite shows a large chromatin nucleus staining intensely, and a small amount of cytoplasm. The nucleus may be centrally or peripherally placed, and is variable in shape. The parasites vary in size from 0·5 to 1·5 microns in diameter and they may be rounded or oval in shape.

In the diplococcus forms there may be a large surface of contact, the contact may be tangential, or the two parts may be quite separate. In such cases one parasite is a little smaller than the other. These forms are no doubt stages in the multiplication of the organism. One, two, three and not rarely four parasites have been observed in a single corpuscle.

The number of red corpuscles invaded by parasites is variable, but in rich specimens 30 per cent. have been found.

Neither *Piroplasma bigeminum* nor blood parasites other than Anaplasma have been found, although numerous specimens have been carefully examined.

Very severe blood lesions are described. In addition to the destruction of red corpuscles the following abnormalities have been discovered: poikilocytosis, the presence of megalocytes, numerous corpuscles showing punctate basophilia, polychromatophilia, and occasional normoblasts.

Conclusions.—

1. There exists in Italy (Roman Campagna) a disease of cattle caused by a particular protozoal parasite—*Anaplasma marginale*.
2. The parasite gives rise to a severe disease characterised by a marked anaemia with important alterations in the blood corpuscles.
3. The infection is apparently transmitted by the *Rhipicephalus bursa*.

(116) Koidzumi (M.). **On the Nature of the "Marginal Points" occurring in the Blood Corpuscles of Cattle.**—*Centralbl. f. Bakt.* 1. Abt., Orig. 1912. July 17. Vol. 65. No. 4-5. pp. 337-340. With 1 plate.

After a preliminary summary of the views expressed by various authors as to the nature of "marginal points" the author passes on to consider the nature of the parasite as it occurs in the blood of cattle in Formosa. It is stated that there is in Formosa an endemic disease of cattle closely allied to Texas fever. Apparently healthy cattle harbour the virus and imported animals are the principal sufferers.

The author states that as a result of his examinations he has been able to collect strong evidence that marginal points are in reality a stage of Babesia, that they persist in the blood of recovered animals for a long time, and serve as the source of infection of imported animals.

Parasites indistinguishable from *Babesia bigemina* appear in the blood of animals suffering from the disease; and when such animals are recovering the marginal points make their appearance in the blood, the large parasites gradually disappearing.

The author states that he has been able to trace several forms intermediate between the two types of the organism. In some cases the parasite assumes the shape of a long-necked flask, the whole of the nuclear substance and the greater part of the cytoplasm being in the rounded part. In others the rounded portion is reduced in size and more dense, the cytoplasm forming merely a tail-like appendage. In still other forms this appendage is reduced to a mere projection.

In the author's opinion the occurrence of what he describes as "large normal forms" and marginal points in the blood at the same time furnishes good reason for believing that the forms above described are intermediate forms.

Marginal points are larger when they have just been formed than in the blood of recovered cattle, and sometimes contain inner structures. Some show a deeply-stained peripheral part with a faintly stained centre, and in others this faintly stained area contains a central intensely stained mass.

Judging from the description given by Sieber the author thinks that *Anaplasma marginale* as it occurs in Africa is different from the parasite observed by him, and he finds himself unable to accept the view that all the "coccus-like bodies" are Anaplasma.

The views expressed by the author in this paper appear to have been based solely on the examination of a number of blood-smears prepared by the Government Veterinary Surgeon in the district.

(117) Schellhase (W.). **Eine Beobachtung über das Vorkommen von Marginalpoints (*Anaplasma marginale*) im Blut von Schafen in Deutsch-Ostafrika.** [The Occurrence of *Anaplasma marginale* in the Blood of the Sheep in German East Africa.] *Berlin. Tierärzt. Wochenschr.* 1912. Vol. 28. No. 28. pp. 511-512.

The author appears to have observed the parasite in the blood of sheep on two occasions only, once in 1909 and once in 1911. The earliest symptoms observed in affected sheep were discharges from the eyes and nose. These were followed by gradual wasting and diarrhoea. There was also intermittent fever and a falling-out of the wool. At the post-mortem nothing characteristic was observed save the lesions typical of anaemia. Parasites (*S. contortus*) were frequently found in the abomasum, but in view of the fact that these were also encountered in healthy sheep the author does not consider that the disease was due to them. The parasite was not found in the early stages of the disease. A very small number of extra-corpuscular organisms were observed, and these were generally very like diplococci in appearance. Transmission experiments could not be undertaken.

(118) Theiler (A.). **Übertragung der Anaplasmosis mittels Zecken.** [Transmission of Anaplasmosis by means of Ticks.] *Zeitschr. f. Infektionskrankh., parasit. Krankh., u. Hyg. d. Haust.* 1912. August 24. Vol. 12. No. 2. pp. 105-116.

1. The transmission of *Babesia bigemina* and *Anaplasma marginale* by means of the larvae of *Boophilus decoloratus* (blue ticks).

Origin of the ticks.—The female ticks were collected from African animals which had constantly been at pasture, and which therefore must have been infected with both *Babesia bigemina* and *Anaplasma marginale*. The larvae were hatched out at the laboratory.

Ox 787.—Imported from England and housed for three months after arrival. This animal had placed upon it about 100 larval ticks. On the 11th and 12th days there was observed an elevation of temperature. Blood examination proved negative. During the following days the temperature was somewhat irregular, but did not go higher than 39·5° C. Babesia was first observed in the blood on the 26th day and again on the 48th day, the temperature being somewhat irregular in the interval. On the 75th day there was a typical febrile reaction, the temperature remaining high from the 85th to the 100th day. Anaplasma first appeared on the third day after the rise of temperature, about 4·5 per cent. of the corpuscles being invaded. The maximum was reached on the 86th day when the percentage was 15·4. The usual lesions of anaemia were observed. After the disappearance of Anaplasma the animal recovered.

Since the usual period of incubation after tick infestation varies from 17 to 25 days the rise of temperature on the 11th day must

be considered as accidental, and if the temperature had not been taken and microscopic examination of the blood made the attack of redwater would have passed unnoticed.

The above experiment shows that a single brood of ticks is capable of transmitting the double infection, but furnishes no evidence as to whether a single tick is capable of harbouring both the parasites. This possibility is, however, not excluded.

2. The transmission of Anaplasma by means of larval blue ticks to English cattle immunised against *Babesia bigemina.*

Experiment A.

Ox 922.—Immunised against redwater in England by a subcutaneous inoculation of 10 cc. of immune redwater blood.

On the 5th day after inoculation the temperature rose to 40·6° C. Blood examination was negative, but the animal was given a dose of trypanblue. The temperature fell rapidly and *Babesia bigemina* was observed for the first time on the 7th day after the initial rise of temperature and again 6 days later. The parasites were present in very small numbers only. The animal was housed directly it arrived in South Africa and observations carried out daily for a long period shewed that its temperature was normal.

The fully-engorged females from which the larvae were obtained were collected from animals in Natal, and since both babesiasis and anaplasmosis occur in Natal it is probable that the animals from which the ticks were obtained were immune to anaplasmosis.

Seventy-seven days after its arrival the animal had placed upon it a large number of larvae. From the 10th to the 14th day there was a slight febrile reaction, Babesia being observed in the blood in very small numbers on one occasion only. Subsequently the temperature was somewhat irregular but examination of the blood proved negative.

On the 55th day there was a further reaction which lasted for 18 days. Anaplasma was observed in very small numbers on the 55th day and two days later 8·9 per cent. of the red corpuscles were invaded. By the 61st day the percentage of invaded cells had risen to 18·8, but after this there was a rapid fall in the number, the percentage being only 0·6 four days later. The parasite was still demonstrable in very small numbers on the 75th day, and appeared to be of the "*centrale*" variety. On the 71st day *Spirochaeta theileri* was observed for the first time and from the 76th to the 86th day there was a second rise of temperature, *Anaplasma marginale* var. *centrale* again appearing in the blood with a maximum invasion of the red corpuscles of 5·4 per cent.

It appears to be possible that the first rise of temperature was caused by the spirochaete and that the appearance of Babesia in the blood was more of the nature of an accident, such as are frequently seen in immune animals.

Experiment B.

Ox 925.—Immunised against redwater in England as in the previous case. The resulting reaction was very mild, parasites being observed in the blood on a number of occasions, but never in large numbers.

The ticks used in this experiment were of the same brood as those used in the previous experiment.

Several thousand ticks were placed on this animal on the 77th day after its arrival, the animal's temperature having been taken daily and found to be normal during the whole of the period.

The first rise of temperature occurred on the 9th day and lasted for a week. Immediately afterwards there was a second rise which was somewhat irregular and lasted till the 30th day. *Babesia bigemina* was found on the 12th day.

From the 45th day there was a third reaction during which the evening temperature rose as high as 41·0° C. Repeated examinations of the blood were made, but no abnormality save slight anisocytosis was discovered. A fourth reaction commenced on the 114th day during which a few Anaplasmata were found which belonged to the "*centrale*" variety.

Possibly in this case also the irregular temperature reactions were referable to the presence of spirochaetes which were overlooked; the possibility is not however excluded that the severe infestation with ticks was responsible. In this case the appearance of Anaplasma was delayed and would no doubt have escaped observation had not the microscopic examinations been continued for so long.

3. The transmission of a pure Anaplasma infection to susceptible cattle by means of ticks (*B. decoloratus*).

Ox 934 was inoculated with blood from a calf that was infected with Anaplasma and *Babesia mutans*. It could be proved in this case that *Babesia bigemina* infection was certainly excluded, as was shown by the typical reactions obtained in both animals when subsequently inoculated with redwater.

The pure Anaplasma infection was obtained by means of blue ticks because it has been proved that this tick is not a host of the *B. mutans*. Ticks free from parasites had to be used in this experiment and these were obtained by collecting the fully engorged females from horses.

The larvae which were hatched out at the laboratory were placed on a clean, freshly imported animal, and if no reaction resulted it might be taken that the ticks from the horses were clean and also that the larvae obtained from uninfected animals must remain clean.

Ox 931 had placed upon it larval blue ticks derived from females that had engorged themselves upon horses. There was no reaction. From this it might be concluded that the ticks obtained from the horse were not infected. The fully engorged females were collected from Ox 931 and the larvae derived from

these were placed on Ox 934 which was infected with Anaplasma and *Babesia mutans*. A large number of ticks were used and from 20-30 days later the fully engorged females were collected. The larvae derived from these ticks could only be infected with Anaplasma.

Calf 1168, which was born at the laboratory and had been kept free from ticks, had placed upon it a number of young larvae obtained from Ox 934. The larvae went through the stages of development and began to drop off on the 24th day. Repeated examinations of the blood proved negative. On the 52nd day there was a febrile reaction which lasted for 18 days, the temperature rising to 40° C. Anaplasma was found in the blood on the day on which the temperature rose. The maximum percentage of corpuscles infected was 7·8 (on the second day of the reaction). From the 8th day to the end of the reaction the number of invaded corpuscles amounted to 1 per cent. The blood lesions of anaemia were pronounced but soon disappeared.

Proof of the purity of the infection.—To furnish proof of the purity of the infection of Calf 1168 blood was drawn from it and injected into Ox 1217 which had arrived about a fortnight previously and which, since arrival, had been housed and kept free of ticks.

A febrile reaction commenced on the 22nd day and lasted till the 31st. Anaplasma was found to be present in the blood from the 22nd to the 44th day, there being also pronounced evidence of anaemia. Neither *Babesia bigemina* nor *B. mutans* was found.

The susceptibility of Ox 1217 to *B. bigemina* was tested by inoculating it with blood from Ox 1216 which had been infected with pure redwater. This resulted in a positive reaction and the animal was treated with trypanblue.

Ox 1218. For sixteen days after its arrival this animal was kept free from ticks and its temperature noted daily without any elevations being observed. It then had placed upon it larval ticks derived from females from Ox 934.

On the 22nd day there was a slight rise of temperature, but blood examination proved negative and the rise was obviously of an accidental nature. Commencing on the 70th day there was a slight but typical febrile reaction which lasted for 15 days. Anaplasma was observed in the blood throughout the reaction and there was evidence of anaemia.

Proof was furnished in the same manner as in the case of Ox 1217 that the infection contracted by Ox 1218 was pure.

The susceptibility of this animal to *B. bigemina* infection was also proved in a similar manner.

4. Transmission experiments with the larvae of *Rhipicephalus simus*.

The fully engorged female ticks were obtained from Natal, and it was presumed that the larvae derived from them were infected either with Babesia or Anaplasma. The ox is not the most favour-

able host for *Rhipicephalus simus* larvae as only a few attach themselves to this animal.

Ox 930.—This animal was housed for a period of three and a half months after its arrival and its temperature noted daily. It then had placed on it a number of *simus* larvae. On the following day it was observed that only a small number had attached themselves.

On the 75th day there was a rise of temperature which lasted until the 100th day, the febrile reaction running a typical course. Anaplasma appeared in the blood at the commencement of the reaction and persisted throughout it. Lesions of anaemia were observed in the blood.

The purity of the infection in this case was tested by using 50 cc. of the blood taken some six months after the animal's recovery and injecting it into a susceptible English animal (1213).

The resulting reaction was a very severe one, the animal suffering from marked anaemia complicated by loss of appetite, inability to stand, and acceleration of respiration. Treatment was resorted to and the animal recovered. Babesia was not observed in the blood during the reaction, but Anaplasma was present in very large numbers.

The susceptibility of Ox 930 to babesiasis was tested by placing on it infected larval blue ticks derived from females engorged on an immune animal. There was a positive reaction, *Babesia bigemina* appearing in the blood from the 29th day. There was a rise of temperature on the 20th day and *Spirochaeta theileri* was found to be present in the blood.

Conclusions.

Five imported English animals and one stall-born Africander calf were used for the experimental transmission of Anaplasma by means of ticks. All the animals were susceptible to the disease. Two of the English animals were immune to redwater the immunity having been conferred experimentally before exportation.

Anaplasmosis was transmitted in every case. In the first experiment both anaplasmosis and babesiasis were transmitted by means of larval blue ticks derived from females engorged on animals immune to redwater and anaplasmosis. In the second experiment the ticks transmitted anaplasmosis to animals immune to redwater. In the third experiment the ticks used were originally obtained from horses and were proved to be entirely free from infection. These ticks were infected by placing them on an animal which had passed through anaplasmosis and *mutans* babesiasis. Only the Anaplasma infection was transmitted as shewn by subsequent inoculations of the English animals with blood. In the fourth experiment it was incidentally proved that the larvae of *Rhipicephalus simus* are capable of transmitting a pure Anaplasma infection. All the animals which passed through an attack of pure anaplasmosis remained susceptible to infection with *Babesia bigemina*.

It must be considered as proved that Anaplasma can be transmitted by means of ticks either by itself or together with *B. bigemina* and *Spirochaeta theileri*. This proves that anaplasmosis and babesiasis are independent conditions. Reference must be made to the comparatively long period of incubation after tick infestation. Full use was made of this fact in the protective inoculation against anaplasmosis (*A. marginale* var. *centrale* against *A. marginale* infection).

Addendum.

The Virus of Anaplasmosis does not pass through a Berkefeld Filter.—In all his investigations the author has considered the rounded bodies contained in the red corpuscles, either centrally or peripherally placed, to be of a parasitic and protozoal nature. This view is based on the staining and biological characters of the bodies.

In spite of the proof furnished in support of this view the idea suggested itself that the so-called Anaplasma might not be parasitic in nature but only a phenomenon associated with the disease—a cell-inclusion such as have been demonstrated in other diseases. If this were the case the virus would be invisible. Since the disease is transmissible by inoculation an answer to the question could be obtained by filtration experiments. With this object the following experiment was undertaken.

A quantity of blood was taken from Ox 1192 which had contracted anaplasmosis naturally and in which 50 per cent. of the corpuscles were invaded. This was diluted with 12 volumes of salt solution and passed through a Nordtmeyer-Berkefeld filter. Three hundred cubic centimetres of the filtrate were inoculated subcutaneously into Ox 1211 (English). There was no febrile reaction, and parasites did not appear in the blood.

Six weeks later the animal was tested with regard to its immunity to redwater and anaplasmosis. It was given 5 cc. of blood from each of the Oxen 1216 and 1212 which were immune to redwater and anaplasmosis.

A febrile reaction resulted and *B. bigemina* was found in the blood on the 7th day. Treatment with trypanblue was resorted to and the parasites disappeared. From the 50th to the 65th day Anaplasma was demonstrable in the blood. The parasite was of the "*centrale*" variety. The reaction was slight.

BABESIASIS.

(119) Pécaud (G.). **La Piroplasmose Bovine au Dahomey.** [Bovine Piroplasmosis in Dahomey].—*Bull. Soc. Path. Exot.* 1912. July. Vol. 5. No. 7. pp. 482-486.

Young animals are principally attacked and in them the disease is benign. Among adult animals only a small proportion are affected, but the disease is far more serious. The principal symptoms are: Weakness, anaemia and cachexia associated with slight inco-ordination, more or less elevation of temperature, diarrhoea, and slight jaundice. Haemoglobinuria is only exceptionally observed. Death takes place in severe cases in fifteen to twenty days.

The lesions found at the post-mortem are not marked. There may be oedema of the lungs with extensive areas of congestion, a large quantity of liquid in the pericardium, petechiae on the heart. The spleen may be enlarged and congested, and in two cases was found to be ruptured. There is enlargement and congestion of the liver. The kidneys may show haemorrhagic

nephritis. The blood is watery and the number of red corpuscles greatly reduced.

The parasite occurs in the blood in four well-marked forms:

(a) Classical bigeminal forms, with, rarely, four parasites in a single corpuscle.

(b) Ring forms having the chromatin collected at one point and a central vacuole.

(c) Rod-shaped and comma-shaped parasites half the substance of which is composed of chromatin.

(d) Small forms arranged in the form of a Maltese cross. The parasites are most abundant at the commencement of and during the acute stage of the disease. They do not disappear completely from the blood, and may reappear if the vitality of the animal is lowered from some other cause.

Details are given of a fatal case which was complicated with ulcerative stomatitis and gastritis.

The following is a summary of a benign case in a calf four and a half months old.

The first symptom observed was weakness of the quarters and slight greenish diarrhoea associated with a little elevation of temperature. For the following five days the condition remained the same, twin piroplasms being very scanty in the blood. Two days later the temperature had risen to 39° C., parasites were numerous in the blood, particularly the twin and ring forms. Punctate basophilia was also observed. The following day piroplasms were rather less numerous. The symptoms gradually abated, piroplasms disappearing from the blood, and the calf appeared to have made a complete recovery two months later. Shortly afterwards the animal was used for the preparation of Jenner's vaccine, and during the period of eruption and for some days after a small number of twin parasites were discoverable in the blood.

Five cubic centimetres of the blood of this calf, withdrawn when the piroplasms were not scanty, were injected into a calf one and a half months old. On the ninth and tenth days the animal appeared to be ill and refused food. There was slight fever. Parasites first appeared in the blood on the nineteenth day, a few twin forms being observed. About a week later ring and rod forms were found. Within ten days the parasites had disappeared and the animal recovered rapidly.

Inoculation experiments have been repeated on two occasions with positive results, every care being taken to avoid the possibility of the results being due to natural infections.

Ticks are very numerous in Dahomey at the commencement of the rains, and the species found are *B. annulatus*, *B. decoloratus* and unidentified species of Amblyomma and Rhipicephalus. Piroplasmosis appears after the rains only (November and December).

Injections of twenty-five to fifty centigrammes of atoxyl cause a rapid decrease in the number of parasites.

THEILERIASIS.

(120) Theiler (A.). **Weitere Beobachtungen, betreffend die Übertragung von Küstenfieber vermittels Zecken.** [Further Observations regarding the Transmission of East Coast Fever by means of Ticks.]—*Zeitschr. f. Infektionskrankh. Parasit. Krankht. u. Hyg. d. Haust.* 1912. Vol. 12. No. 1. pp. 26-42.

As is well known the parasite of East Coast fever can be transmitted by five species of ticks, viz., *Rhipicephalus appendiculatus*, *R. evertsi*, *R. simus*, *R. nitens*, and *R. capensis*.

The species which occurs most frequently in countries where the disease is known is the *R. appendiculatus*, and it was with this tick that the author's experiments were carried out.

Experiment 1.

A. *Brown ticks which have been infected with East Coast fever as larvae, transmit the disease to susceptible animals in the nymph stage, but are non-infective in the adult stage.*

R. appendiculatus Imago (No. 268).

Fully engorged female ticks, obtained from Natal, deposited eggs a few days after reception, and these hatched out a month later.

Larvae placed on an infected animal for three days when the parasites were numerous in the blood.

Larvae moulted normally and the resulting nymphs were placed on a susceptible animal. The disease developed on the 15th day, and 12 days later the animal was slaughtered for inoculation experiments. The nymphs engorged themselves, and after moulting were placed upon two other animals.

(b) *R. appendiculatus* Imago (No. 309).

Larvae derived from fully engorged females were placed on calf 700. They engorged themselves and dropped off at a time when the parasites were numerous in the blood. As nymphs they were placed on calf 917 and the animal developed the disease and died; parasites being numerous in the blood and agamonts and gamonts in the glands. The females developed from nymphs collected from this calf were placed upon two other animals (1145 and 1088), in order to test their infectivity.

(c) *R. appendiculatus* (No. 335).

Larvae placed upon calf 917 when suffering from the disease, the ticks being collected at a time when there were numerous parasites in the blood.

The resulting nymphs were used together with those from animals 700 and 923 (ticks 268 and 309) on eight animals (Nos. 561, 908, 919, 1011, 914, 1012, 1026, and 1040).

Six of the eight animals contracted the disease.

B. *Brown ticks which have been infected with East Coast fever as larvae are uninfective for animals that are immune to the disease as a result of inoculation, and are cleansed by being placed upon such animals.*

Larvae Nos. 309, 268, and 335 were fed upon infected animals and proved to be infective in their nymphal stage by placing them upon susceptible animals, but nymphs derived from exactly the same source proved to be non-infective when placed upon 10 animals that had been rendered immune to the disease experimentally.

C. *Ticks which have passed their nymphal stage either upon susceptible animals to which they have transmitted the disease or upon immunised animals fail to transmit the disease to susceptible animals in the adult stage.*

Six experiments of this nature were carried out with negative results.

The author draws the following conclusions from this series of experiments:—

Ticks which fed as larvae on diseased animals and transmitted the disease as nymphs to eight susceptible animals, or which fed as nymphs on twelve immunised animals, failed to transmit the disease when placed in larger numbers on six susceptible animals.

A number of animals were used in the experiment and a large number of ticks were employed so that the possibility of any accident can be excluded.

Experiment 2.

Ticks which have fed upon animals suffering from East Coast fever do not always transmit the disease in the succeeding stage.

Details regarding the ticks used.

1. *R. appendiculatus* No. 363.

Larvae placed upon ox 1013 on the 7th day after the rise of temperature, and collected eight days later; this being three days before the animal died from the disease. Parasites were numerous in the blood and all the developmental stages of the parasite were discovered in the glands.

2. *R. appendiculatus* No. 364.

These ticks were placed upon an infected animal (No. 908) on the 5th day of the disease and fed for 10 days.

3. *R. appendiculatus* No. 355.

The larvae fed for 5-7 days upon an infected animal (No. 913).

4. *R. appendiculatus* No. 356.

Fed for 3-6 days upon animal 914. This animal recovered from the disease.

5. *R. appendiculatus* No. 349.

Fully engorged nymphs were taken from a diseased animal in Natal and moulted in the laboratory.

6. *R. appendiculatus* No. 411.

Fed both as larvae and as nymphs on affected animals (Nos. 914 and 1053).

7. *R. appendiculatus* No. 426.

Larvae hatched out from eggs laid by engorged females obtained from Natal. Fed upon a susceptible animal and after moulting were fed upon Ox 1111 which was suffering from East Coast fever.

8. *R. appendiculatus* No. 373.

The mature females were obtained from animals 906, 1088, 1009, and 1021. The larvae were placed on ox 909 when it was the subject of the disease.

TESTS OF THE INFECTIVITY OF THE TICKS.

(a) *Brown nymphs* 363, 364, 355, 356, *and* 349.

1. Ox 627.—This animal had previously been used for inoculation experiments in connection with the transmission of the disease with negative results.

On three separate occasions this animal had placed on it a number of nymphs (Nos. 363, 364, 355, and 356), but in no case was there any reaction.

Subsequently there were placed upon it 6 imagines obtained from Natal. Three of these attached themselves. The animal developed the disease which terminated fatally.

2. Ox 911.—This animal had also been used in transmission experiments with negative results.

On two occasions this animal had placed on it 20 nymphs (Nos. 364, 355 and 356) without there being any reaction.

Six brown ticks from Natal were subsequently placed upon it with the result that a fatal attack of the disease was set up.

3. Ox 1014.—The history of this animal was the same as of the preceding ones save that nymphs failed to set up the disease on four separate occasions.

4. Ox 1068.—History like that of No. 1.

(b) *Brown Nymphs Nos.* 363 *and* 373.

5. Ox 1037.—Had been used in experimental work without result. Twenty-seven ticks (No. 363) failed to cause any reaction. Five months later 20 nymphs from ox 909 (No. 373) caused a fatal reaction.

(c) *Brown Nymphs Nos.* 363, 364, 349, 411, *and* 426.

6. Ox 1046.—Animal had been used unsuccessfully in transmission experiments.

On two occasions nymphs and on one occasion adults from Natal were placed upon this animal without result.

Subsequently a fatal attack of the disease was set up by 10 adults, 8 (No. 411) from animal 1053, and 2 (No. 426) from ox 1111. Only five of the ticks actually attached themselves.

(d) *Brown Nymphs* Nos. 363 and 364.

7. Ox 1043.—Previous history as before.

First batch of ticks (nymphs No. 364) failed to set up the disease.

Six weeks later a second batch of the same ticks set up a fatal attack.

8. Ox 1017. As ox 1043, save that the ticks which failed to set up the disease were No. 363, and those which caused the fatal attack were No. 364.

(e) *R. appendiculatus* (Nos. 364, 349, 411, and 426).

9. Ox 1090.—Had not been used before. Was obtained from a district free of the disease.

Ten ticks No. 364 were placed upon the animal but only 6 attached themselves. There was no reaction.

A month later four adult ticks from Natal were placed upon the animal, two attached themselves without producing any reaction.

Three weeks later 6 adults (No. 411) from animal 1053 were used, five of which attached themselves.

A week later two more of the same ticks were put on the animal and also two from animal 1111 (No. 426). The animal contracted the disease and died.

(f) *R. appendiculatus* Nos. 363, 364, and 349.

10. Ox 1082.—This animal was obtained from a district free of the disease and had not been used for experiment.

On two separate occasions brown nymphs (Nos. 363 and 364 respectively) were placed upon this animal without producing a reaction.

Subsequently a reaction which terminated fatally was obtained by placing 6 adult ticks (No. 349) obtained from Natal on the animal.

(g) *R. appendiculatus* nymphs No. 364.

11. Ox 1050.—This animal had been used previously for a transmission experiment which was unsuccessful; infection and death followed the attachment of 7 nymphs (No. 364).

From these experiments it may be seen that;

Ticks No. 363 were not infected.

Not all the ticks in batch 364 were infective.

Batches 355 and 356 were not infected.

Batch No. 349 were infective, but in two cases in which two and three ticks were used the disease was not transmitted.

Batch No. 373 were infective.

Batches 411 and 426 were infective.

Experiment 3.

Adult brown ticks which fed as larvae on infected animals and were virulent as nymphs failed to transmit the disease when the nymph stage was passed upon rabbits.

1. *R. appendiculatus* No. 342.

The larvae were derived from adults collected in Natal and were placed upon ox 923 at a time when parasites were numerous in its blood. The engorged larvae were collected and moulted normally. A number of them were placed on animal 596 and transmitted the disease, others were placed upon a rabbit, the engorged nymphs being collected.

2. *R. appendiculatus* No. 309.

Larvae derived from adults collected in Natal were placed on calf 700 which was suffering from the disease. After moulting some were placed on calf 917 and transmitted the disease, and others were placed on a rabbit.

Test of the virulence of the Ticks.

1. Ticks from both of the rabbits were placed on ox 1059 in succession without producing the disease. The animal subsequently died from East Coast fever.

2. Ox 1019 had three batches of ticks from the rabbits placed upon it, two from the first and one from the second, without any reaction being caused. The animal subsequently died from the disease naturally contracted.

Experiment 4.

Adult ticks which have passed their nymph stage on animals which have recovered from East Coast fever are unable to transmit the disease to susceptible animals.

The immune animal used in this experiment was No. 914. This ox was infected by nymphs Nos. 268, 335, and 309. Blue bodies were demonstrable in its glands, and *T. parva* was present in its blood. The animal recovered.

The ticks which were placed upon the immune animal were nymphs No. 298 obtained from ox 868.

Test of the Ticks.

The engorged nymphs were collected and after moulting were placed on ox 1021. There was no reaction.

Calf 1130 also had some of the ticks placed on it but without any subsequent reaction.

Conclusions.

1. Brown adults which were infected as larvae, and which transmitted the disease as nymphs, are not infective for susceptible animals. Three different batches of ticks were used. In the nymph stage they transmitted the disease to eight animals, but failed to transmit it to two susceptible animals in the adult stage.

2. Ticks belonging to the same batches which had reached the nymph stage on animals experimentally immunised against East Coast fever failed to transmit the disease in six cases in the adult stage. This proves that brown ticks which become infected in one stage are cleansed in the following stage whether they engorge themselves on immune or susceptible animals.

3. Ticks infected as larvae which passed the nymph stage on rabbits were not infective for susceptible animals as adults. This confirms the foregoing fact that a tick loses its infection the first time it feeds on a susceptible or immune animal.

4. Clean or infected ticks which feed on a recovered (immune) animal do not transmit the disease in their next stage. This conclusion was arrived at eight years ago. (See *Annual Report of the Government Veterinary Bacteriologist, Transvaal Dept. of Agriculture*, 1904–1905.)

5. It has been shown that different batches of ticks which were collected at the same time and kept under the same conditions do not transmit the disease during the next stage even when placed upon animals in large numbers. Other batches kept under the same conditions and collected in the same way infect a few animals only, while still others prove to be infective on every occasion, and even when used in small numbers only.

It is difficult to explain this fact but it is quite possible that external circumstances have an influence in some way or other. Those ticks which failed to transmit the disease were fed during the coldest periods of the year. This is possibly the explanation of the fact that the disease in the field is relatively less severe during the winter months; but the fact should not be lost sight of that at this period of the year the ticks are not particularly active.

TRYPANOSOMIASIS.

(121) Blacklock (B.). **The Trypanosomes found in a Horse Naturally Infected in the Gambia. A Double Infection.**—*Ann. Trop. Med. & Parasit.* 1912. May 29. Vol. 6. No. 1. B. pp. 107-116.

In the first place reference is made to the conclusion of a previous paper* published in 1911, the reference being as follows:—

"A former paper (1911) dealt with the trypanosomes found in two horses naturally infected in the Gambia. These horses were referred to as *Horse A* and *Horse B*. The former, *Horse A*, contained in its peripheral blood two forms of parasite, one long the other short; the latter, *Horse B*, presented one form of parasite only. The conclusion of the paper was as follows:—

"(1) We consider the trypanosome found in *Horse B* to be *T. dimorphon, sensu* Laveran and Mesnil.

"(2) The long form in *Horse A* appears to us to be *T. vivax*.

"(3) As regards the short form found in *Horse A*, we do not feel justified at the present stage in assigning its position. It may be a Dimorphon-like trypanosome of low pathogenicity, or simply a modification of the long parasite of *Horse A*."

The two forms of trypanosomes were separated by animal inoculation and from this the author adduces evidence of the dual infection. Apart from the morphological differences the following evidence proves this duality.

The long form of trypanosome has been kept running for a year in goats, maintaining its morphology for the whole of the time,

* Yorke & Blacklock. The Trypanosomes found in Two Horses naturally infected in the Gambia. *Ann. Trop. Med. & Parasit.* 1911. Vol. 5. No. 3. p. 413.

and short forms have never appeared in any goat save in one instance at the first passage. The short form was in the minority in the proportion of about one to a thousand of the long forms. In monkeys, guinea-pigs, rabbits, rats, mice, and dogs only the short form has appeared, save in one rabbit of the first passage (inoculated from the horse) in which long forms predominated till the last day, when the short forms suddenly became numerous. The short form after being carried through a number of smaller animals was inoculated back into a horse. The blood of this animal was found to be highly infective for a goat, dogs, rabbits, guinea-pigs, rats, and mice, but only the short forms appeared.

Many laboratory animals having proved themselves refractory to the long form were readily infected with the short form.

A small number of laboratory animals inoculated intraperitoneally from goats infected with the long form became infected, and parasites were found in their blood for brief periods, but the strain could not be carried on in the same species.

Pathogenicity of T. vivax *in goats.*—The average period of incubation was nine days, and the period of illness thirty-one days. Every goat inoculated became infected, and none recovered. There was no marked diminution in the period of incubation or in the duration of the disease.

It was observed that *T. vivax* after it had been preserved for some time in goats gradually became slower and slower in its movements. This change of movement commenced in October and persisted till March, when the original motility returned. Circumstances indicated that this slackening of the movement was not due to conditions of temperature in the laboratory.

Two strains of short forms were kept running, one in rats only derived from the horse inoculated from a small laboratory animal, and one in rabbits derived from the original horse. There was observed to be a marked increase in virulence of the strain passed through the horse, the period of incubation being reduced from eighteen to four days.

(122) Bruce (Sir D.), Harvey (D.), Hamerton (A. E.), Davey (J. B.), & Lady Bruce. **The Morphology of** *Trypanosoma simiae*, **sp. nov.**—*Proc. Roy. Soc.* 1912. August 24. Series B. Vol. 85. No. B. 581. pp. 477-481. With 1 plate and 2 text-figures.

This trypanosome is remarkable in that it attacks only such widely different animals as the monkey and the goat. Oxen, baboons, dogs, guinea-pigs and white rats appear to be immune. The trypanosome is fatal with great rapidity to monkeys. In a series of nineteen cases the average duration of life after the trypanosomes were first seen in the blood was only 2·9 days. In the Kasu Hill district the carrier is *Glossina morsitans*.

In the living unstained state the trypanosome shows active translatory movements. In preparations fixed with osmic acid and stained by Giemsa the measurements of the trypanosome were found to be as follows. In the monkey the average length was

18·1 microns, the maximum being 24 and the minimum 14. In the goat the average length of the trypanosome was about one micron less than in the monkey, the maximum being 21 and the minimum 14.

The species is monomorphic. The average breadth is 1·75 microns, the maximum being 2·75 and the minimum 1. The body is frequently extended in a straight line. The posterior extremity is blunted, pointed or rounded, and there is frequently the appearance of a vacuole at the extreme end. The body substance is free from granules. The nucleus is oval and situated about the middle of the body. The micronucleus is small and round, situated about 1½ microns from the posterior end of the body. A peculiarity is that it is always placed at the edge of the trypanosome and appears to protrude a little. The trypanosome differs from the *T. vivax* and *T. uniforme* in that the undulating membrane is well developed. It is difficult to say whether the species has a free flagellum or not.

In the monkey these trypanosomes swarm in enormous numbers in the blood. Large masses of them can sometimes be seen, the masses being sufficiently large to fill up the whole field of the microscope. It would appear as if multiplication took place so rapidly that individual trypanosomes had not time to disengage themselves. Other division forms are observed in which the trypanosomes appear to be joined together by their non-flagellar ends.

"*Conclusions.* (1). *T. simiae* is a well-defined species, easily separated by its morphology alone from the other trypanosomes which have been described as causing disease among domesticated animals.

"(2) It sets up a chronic disease in goats, but is chiefly remarkable for its rapidly fatal action on monkeys.

"(3) In Nyasaland it is carried by *G. morsitans* and in this district—Central Angoniland—this tsetse-fly is found to be heavily infected with this trypanosome."

(123) Kinghorn (Allan) & Yorke (Warrington). **Trypanosomes obtained by feeding Wild *Glossina morsitans* on Monkeys in the Luangwa Valley, Northern Rhodesia.**—*Ann. Trop. Med. & Parasit.* 1912. Sept. 12. Vol. 6. No. 3. A. pp. 317-325.

Unfortunately owing to the lack of healthy goats and sheep these animals could not be used in the experiments with wild *G. morsitans*, monkeys being the only animals available for the purpose. In spite of this a certain amount of evidence exists that both *T. vivax* and *T. nanum* are naturally transmitted by this fly, although proof is not forthcoming.

In the first series of experiments 3,008 flies were fed on 28 monkeys. Nineteen of the monkeys became infected. The trypanosomes isolated were the *T. rhodesiense*, *T. pecorum* and a third hitherto undescribed parasite for which the name *T. ignotum* is suggested. The proportion of wild fly infected with these trypanosomes is given as follows: *T. ignotum* 1:300, *T. rhodesiense* 1:534, *T. pecorum* 1:1600.

T. ignotum.—This parasite was obtained in ten experiments.

Morphology.—In fresh preparations this parasite appears as a comparatively short and slender organism. It is fairly actively motile but exhibits no marked degree of translatory power. In stained preparations it is observed that the trypanosome is slender, the posterior extremity is obtuse or bluntly rounded, the nucleus is rounded or oval and lies at the middle of the body. The centrosome is small and while usually situated near the posterior extremity it may be separated from it by an appreciable interval. The undulating membrane is feebly developed and a short free flagellum is only occasionally seen. The cytoplasm shows no granules or vacuoles. The average length of 200 individuals was 17 microns, the maximum 23, and the minimum 12.

Pathogenicity.—The virulence of the trypanosome is very great for monkeys whether the infection be through the agency of flies or by direct transmission from monkey to monkey. The average period of incubation was seven days and death occurred 2 or 3 days after the parasites appeared in the peripheral blood. A rabbit was successfully infected by inoculation from a monkey, the period of incubation was 22 days and the duration of the disease 66. One guinea-pig, five rats, and four mice were found to be refractory and negative results followed the feeding of infective flies upon rats.

Diagnosis.—*T. ignotum* most closely resembles *T. uniforme* but it is easily distinguished from that organism by the absence of a free flagellum and also by differences in the pathogenic range. Although the parasite has been isolated from wild *G. morsitans* more frequently than any other trypanosome it has never been found in game or domesticated stock and no information is at present obtainable regarding its original host.

Transmission.—In one experiment the infective fly was determined to be one of a group of ten. These were then killed and dissected. In nine of them no trypanosomes were found, while in the tenth a heavy infection of the proboscis was encountered. The gut, salivary glands and sucking stomach were negative.

(124) Kinghorn (Allan) & Yorke (Warrington). **Trypanosomes infecting Game and Domestic Stock in the Luangwa Valley, North-Eastern Rhodesia.**—*Ann. Trop. Med. & Parasit.* 1912. Sept. 12. Vol. 6. No. 3. A. pp. 301-315.

Glossina morsitans is everywhere abundant in this valley, but in spite of that a large and varied selection of game is found, and probably because of that domestic stock is very scarce.

The authors found it advantageous to examine the blood in preparations dry-fixed in alcohol and stained by Giesma rather than to examine moist preparations owing to the rapidity with which the trypanosomes degenerated. The smears were made from blood from one of the arteries immediately after the animals were shot.

Owing to the impossibility of getting "clean" sheep and goats all the inoculations from game were made into monkeys and rats. The authors recommend, however, that when possible sheep and goats should be used as well since they are susceptible to most of

the pathogenic trypanosomes. This is important when dealing with the *T. vivax* and *T. nanum*.

One hundred and twenty-seven head of game, comprising nineteen genera, were examined, and trypanosomes were found by direct examination, by inoculation, or by both methods in thirty-three. Direct examination was successful in twenty-six cases. Had a number of smears been examined from each animal this number might possibly have been higher. In several instances only a single trypanosome was found in a smear covering the greater part of a slide.

Inoculation experiments showed that 37·5 per cent. of the animals were infected with trypanosomes. Both *T. vivax* and *T. nanum* were found in game, but no conclusions could be drawn as to the presence or absence of these parasites in animals in which they were not found microscopically, since the animals used for the inoculation experiments were not susceptible to either of them.

Different species of animals appear to differ widely in their susceptibility. Trypanosomes were never found in zebra, wildebeest, or bushpig. Waterbuck, bushbuck and kudu were found to be the most heavily infected species, the percentage in these varying from 66·6 to 57·1.

These differences may possibly be referable to some extent to differences of habit of the species, the most seriously infected species being found in thick cover where they would be more exposed to the bites of flies.

A table is given showing the species of trypanosome found in each of the infected animals. In compiling the table the information obtained from the results of inoculation, where that was resorted to, has been utilised. In this way the differentiation between *T. pecorum* and *T. nanum* can be established, these parasites being indistinguishable morphologically. Double infections were not uncommon.

As all the animals shot appeared to be in perfect condition and presented no symptoms of disease, it is probable that their tolerance of the trypanosomes is great, and especially as they have increased in numbers since rinderpest swept through the country.

Examination of domestic stock.—Cattle were found in one village only, and there were only three animals. Two of them appeared to be in good condition, but the third was obviously diseased. These animals represented the remnant of a large herd that had existed there some four or five years before.

The cow in which trypanosomes were found was bred on the Government Farm and had never been outside the limits of the township. Tsetse flies had never been seen within some miles of the place, but Stomoxys was abundant in the kraals, and, at certain seasons, various species of Tabanidae were common.

A few goats were seen, but these were all dead a few months after.

Glossina morsitans was found around all these villages.

Four goats were under observation for some length of time, but trypanosomes were found in the peripheral blood on rare occasions only.

A dog was found infected with *T. rhodesiense*. This animal came from just over the Nyasaland border. The natives said that it had not been out of the village for over a year, and, as the disease in this species runs a very rapid course, there is no doubt that it was infected locally.

The authors give a brief description of the morphology, pathogenicity, and transmission of the trypanosomes found from which the following details are taken.

T. rhodesiense was found in 16 per cent. of the animals from which inoculations were made.

T. vivax was found in eight waterbuck, one kudu, and three goats. Monkeys, rabbits and rats were found to be insusceptible. The parasite is probably transmitted by *G. morsitans*.

T. nanum.—This trypanosome was found in one bushbuck, three waterbuck, and two goats. It was possibly present in other animals but, in the absence of inoculations, this point could not be settled, as the parasite is not distinguishable from *T. pecorum* morphologically.

The trypanosome is sluggish in its movements and there is not much translation. Inoculation failed in three monkeys, one rabbit, and three rats. There is some evidence to show that it is transmitted by *G. morsitans*.

T. pecorum was found in one bushbuck, one mpala, four waterbuck, two kudu, three dogs, and one wild rat.

Presumably all the animals inoculated—nine monkeys, one rabbit, one guinea-pig, eleven rats, and two mice—became infected.

Evidence was obtained in one case that *G. morsitans* is capable of transmitting this trypanosome, but there is a large amount of evidence that other carriers, and probably more important ones, exist.

Evidence was obtained in one case that possibly Tabanidae are capable of transmitting the infection.

In support of the view that flies belonging to this genus are capable of transmitting trypanosomes it may be mentioned that within the past four years there have been three outbreaks of trypanosomiasis among cattle in districts where tsetse flies may be absolutely excluded. Stomoxys and Lyperosia are constantly present, and during the rains various species of Tabanus, Pangonia, and Haematopota are common.

A trypanosome which has not yet been identified was found in one bushbuck.

In fresh preparations the parasite was markedly polymorphic. In stained preparations the trypanosome generally resembled the *brucei* or *gambiense*, except for the presence of occasional *pecorum*-like forms. The maximum length was 33·25 microns, the minimum 10·75, and the average 20. Monkeys, rabbits and rats were infected experimentally, but a guinea-pig failed to become infected.

A second trypanosome, the species of which was not known, was found in a dog obtained from the hills on the Nyasaland border.

The parasite in moist preparations appeared as a broad stumpy organism which had no degree of translatory power.

In stained preparations the following details could be made out. The trypanosome was very broad in comparison to its length, the ratio of breadth to length being 1:4·8. The nucleus was situated at the middle of the body or posterior to it. The blepharoplast was very large and rounded, and situated near the posterior end. Frequently it lay at one edge of the parasite and projected as a small excrescence. The undulating membrane was poorly developed or absent. The cytoplasm contained coarse granules and vacuoles. The average length was 15·8 microns, with a maximum of 20 and a minimum of 10.

A rat showed no trypanosomes up to the thirteenth day after inoculation from the dog, and then it was accidentally killed. The dog died some days previously. The strain was thus lost.

The authors suggest that possibly the trypanosome was the same as that described by Montgomery and Kinghorn in 1909, for which Laveran suggested the name *T. montgomeryi*.

Conclusions.

"1. Trypanosomes are of frequent occurrence in game and domestic stock in the Luangwa Valley. At least 37·5 per cent. of the buck harbour parasites.

"2. Six species of trypanosomes were found, viz.: *Tryp. rhodesiense*, *Tryp. vivax*, *Tryp. nanum*, *Tryp. pecorum*, and two others, of which one was possibly *Tryp. montgomeryi*.

"3. *Glossina morsitans*, in nature, transmits two of these trypanosomes, viz.: *Tryp. rhodesiense* and *Tryp. pecorum*, and probably also transmits at least two others, namely, *Tryp. vivax* and *Tryp. nanum*.

"4. Circumstantial evidence exists to shew that *Tryp. pecorum* may be transmitted by biting insects other than tsetse flies."

(125) Laveran (A.). **Au Sujet du** *Trypanosoma pecorum*.—*Bull. Soc. Path. Exot.* 1912. June. Vol. 5. No. 6. pp. 372-375.

The author has already expressed his opinion that the *T. pecorum* is not identical with the *T. congolense* or *T. dimorphon*, basing his view upon the following facts: 1 The serum of a goat which was immune to *T. congolense* and *T. dimorphon* and which was active against these two viruses, was proved to be inactive when mixed with blood containing *T. pecorum*. 2 A goat immune to *T. congolense* and *T. dimorphon* became infected when inoculated with *T. pecorum*.

The author adds a number of fresh details which give additional support to his view. The goat referred to died from the infection, and at the post-mortem the following lesions were found. The goat was only a little more than half the weight that it was three months before death. The inguinal and axillary glands were enlarged. The spleen weighed 90 grammes. No other gross lesions were discoverable. There was severe dry arthritis principally involving the knees, but this lesion ante-dated the inoculation with the trypanosome.

An African sheep which had been successively inoculated with *T. pecaudi*, *T. dimorphon*, and *T. congolense*, and which had acquired immunity to all three was inoculated with blood from a guinea-pig containing the *T. pecorum*. Trypanosomes appeared

in the blood in very small numbers a fortnight later, after which they disappeared from the blood for some time. A dog which was inoculated intraperitoneally with thirty cubic centimetres of blood from the sheep six weeks after trypanosomes had appeared in its blood became infected and died in eleven days. A second dog inoculated in the same manner four months later also became infected and died. A month or two later the sheep began to show signs of ill health and gradually wasted. Death took place nine months after inoculation. The inguinal glands were somewhat enlarged, the peritoneal cavity contained a small quantity of serous liquid, the spleen was enormously enlarged and weighed 820 grammes, and all the other organs appeared normal.

A second goat inoculated with guinea-pig blood containing large numbers of *T. pecorum* became infected on the 10th day. The parasites were present for six days and then disappeared. During the next ten months six dogs were inoculated at intervals and all of them became infected. A seventh dog inoculated two months later failed to become infected.

Two months later the goat was reinoculated with a large dose of guinea-pig blood containing the trypanosome.

A dog inoculated from it three weeks later failed to become infected.

After a further interval of two months the goat was inoculated with blood containing the *T. congolense*, infection resulted and trypanosomes were found in the blood eight days later. Trypanosomes were found in small numbers in the blood subsequently and the temperature varied a little. Up to the time of writing the infection had not proved fatal, but the author states that the reaction may be considered to be exactly like that which would have been obtained with a goat that had not been infected with the other trypanosome.

(126) Bouffard (G.). **Quelques Considerations d'Ordre Prophylactique concernant le** *Trypanosoma cazalboui*. [Prophylactic Measures against *Trypanosoma cazalboui*.]—*Bull. Soc. Path. Exot.* 1912. June. Vol. 5. No. 6. pp. 380-385.

Trypanosoma cazalboui is the most widely spread animal trypanosome in the West African French Colonies. Together with *T. dimorphon* it renders whole tracts of country uninhabitable for equines and bovines. *T. pecaudi* is to be found along the banks of all the rivers where they are wooded. Whereas *T. pecaudi* is the cause of a sub-acute disease in the horse, is apparently harmless for the donkey, and produces a chronic disease in the ox, *T. cazalboui*, on the other hand, is very virulent for the ox, causing death in as short a period as eight days, but more generally proving fatal in about six weeks.

In the horse it is responsible for a disease which develops very slowly and may last for two to four years. All the Soudanese species are very susceptible.

The disease is rarely encountered in the sheep, but the species is very susceptible to inoculation. The wool-breed in the neighbourhood of Timbuctoo is particularly easy to infect, trypanosomes being very numerous in the blood by the eighth day.

Diagnosis is not associated with difficulty because trypanosomes are always to be found in the blood in eight days, and the morphological characters, and in particular the extreme activity, are sufficiently characteristic. Goats are less susceptible than the sheep, but they are easily infected and are considered by the author to be the " touchstone " in the diagnosis of Souma.

Probably the *G. palpalis* and *G. tachinoides* are the principal agents in the transmission of the disease, but the author has shown that a Stomoxys, although not capable of transmitting the disease in the cyclical manner, is by no means a negligible factor. Probably this fly is responsible for the formation of endemic zones outside tsetse areas. These endemic zones are so to speak of an accidental nature and depend upon the introduction of a diseased animal into a stable, the flies acting as carriers to neighbouring animals. These zones may remain infected for long periods provided fresh animals are being constantly introduced into the stables.

The possibility of the existence of persistent centres of Souma should result in a careful examination of all animals moving from place to place. The routes traversed should be provided at intervals with rest houses sufficiently removed from the local animals to obviate the possibility of direct transmission.

The danger is no less great for horses, and the author has seen losses in areas free from tsetse among stallions imported from France.

On several occasions the author has used the sheep as a test animal for inoculation with blood from horses in which the trypanosome could not be found by direct examination, and with considerable success.

The disease is more rapidly fatal in European horses than in the native races, but even in these it lasts ten months, and the trypanosome cannot always be found in the blood.

According to the author the only method of dealing with the disease satisfactorily in breeding stables is to slaughter every animal that is proved to be infected by the inoculation of its blood into a sheep.

The author has made experiments with atoxyl, the benzidine dyes, orpiment, and arsenophenylglycin but has not had a single encouraging result. In every case in which there has been a disappearance of the trypanosome from the circulation there has always been a relapse.

Every horse coming from a tsetse district should be isolated until its blood has been tested experimentally on a sheep. The absence of tsetse is often wrongly interpreted. It by no means indicates an absence of trypanosomaisis, and it may produce a false sense of security.

(127) Bouet (G.) & Roubaud (E.). **Expériences de Transmission des Trypanosomiases animales de l'Afrique Occidentale française par les Stomoxes.** [Experimental Transmission of Animal Trypanosomes in French West Africa by Stomoxys.]—*Bull. Soc. Path. Exot.* 1912. July. Vol. 5. No. 7. pp. 544-550.

a. *Trypanosoma cazalboui.*

Experiment 1. A healthy kid was kept beside an infected goat in the open from the 15th September till the 1st October. The ears and the flanks of both animals were clipped in order to facilitate biting of the flies. The kid did not become infected.

Experiment 2. Sixty Stomoxys (*S. calcitrans, bouvieri,* and *boueti*) kept in two cages were allowed to bite alternatively and without interval from 28th August to 1st September one infected sheep and one healthy sheep. Trypanosomes appeared in the blood of the sheep on the 2nd September.

b. *Trypanosoma pecaudi.*

Experiment 3. A healthy young kid was kept at liberty with an infected kid from 14th September to 1st October. The ears and flanks of both animals were clipped. Flies constantly tormented them. The healthy kid did not become infected.

Experiment 4. Two healthy puppies were kept at liberty with an infected dog and close to some infected cats from 15th September till 29th October. Neither of the dogs became infected in spite of repeated bites.

Experiment 5. A healthy cat was kept at liberty with an infected cat from the 16th to the 22nd October. The cat did not become infected.

Experiment 6. A healthy dog kept with an infected dog for 10 days and after an interval of a fortnight for a further month with another infected dog failed to become infected.

Experiment 7. Five Stomoxys (*calcitrans*) were fed alternately and without interval on an infected guinea-pig and a healthy pig. After the last feed on the infected guinea-pig they were transferred with an interval of five minutes to a healthy guinea-pig. This experiment was repeated daily for six days but neither the pig nor the guinea-pig became infected.

Experiment 8. One hundred Stomoxys (*S. calcitrans, soudanense, bouvieri, glauca*) kept in two cages were fed alternately on a healthy and an infected cat every day for six days. Trypanosomes appeared in the blood of the previously healthy cat the day after the last feeding.

c. *Trypanosoma dimorphon.*

Experiment 9. A healthy kid was kept with an infected goat for four and a half months in the open and was constantly bitten by Stomoxys. The kid failed to become infected.

Experiment 10. One hundred Stomoxys (*calcitrans*) in cages were fed alternately on an infected goat and a healthy goat for five days. The healthy goat did not become infected.

These experiments show that the transmission of these trypanosomes by Stomoxys is nothing like so easy under natural circumstances as would appear from the results obtained when cages are used. Even when cages are used there is a certain amount of variation with regard to transmission of the trypanosomes.

Comparing these results with the following experiments carried out with the Sahara virus the role of the Stomoxys in the transmission of virus naturally transmitted by Glossina is obviously a secondary one.

Transmission of the Saharan Viruses by Stomoxys.

a. *Trypanosoma soudanense.*

The virus used was of the Tahaga type and was obtained from the valley of the Niger.

1. Transmission by immediately successive bites.

Experiment I. A hundred flies (*S. calcitrans*, var. *soudanense*, *S. bouvieri*, and *S. vittata*) were fed alternately without interval on an infected dog and a healthy rat for two days. The rat became infected a fortnight later.

Experiment II. Similar to Experiment I. with corresponding result.

Experiment III. Fifty flies fed alternately on an infected dog and a healthy guinea-pig with an interval of half an hour. The guinea-pig died a week later without showing trypanosomes.

Experiment IV. Repetition of Experiment III. using an infected rat in the place of the dog. The guinea-pig was not infected a month later.

Experiment V. Flies fed alternately without interval on an infected rat and a healthy dog for three days. The dog became infected a week later.

Experiment VI. Repetition of Experiment V using an infected dog in the place of the rat. The healthy dog was found to be infected a week later.

2. Natural mechanical transmission.

Experiment VII. A young healthy dog was freed from fleas, washed with cresyl, and kept during the day in the open with an infected dog. During the night the animal was placed in a fly-proof cage. The dogs were kept in contact for eight days. On the sixth day after the separation of the two animals the healthy dog showed trypanosomes.

3. Transmission after an interval of at least 24 hours.

Experiment VIII. About eighty of the flies used in the previous experiment were allowed to bite a healthy dog for six days commencing one day after they had had their last feed on the infected dog used in experiment VII. The dog was found to be infected a month later.

The dog was examined up to the end of the following month, but trypanosomes were not discoverable microscopically in its blood until two days before its death. A month before death occurred the serum of this animal if left standing at the temperature of the laboratory became a rich culture of the trypanosomes within a few hours.

A second experiment of this kind in which the infected flies were kept for forty-eight hours before being placed on the healthy animal was abortive owing to the escape of the dog.

b. *Trypanosoma evansi.*

In a series of twelve experiments exactly analogous results were obtained with this trypanosome.

(128) Brieger (L.) & Krause (M.). **Chemotherapie bei Trypanosomeninfektion** (*Trypanosoma brucei*) **nach Verabreichung per os.** [The Treatment of Nagana by means of Drugs administered by the Mouth.]—*Berlin Klin. Wochenschr.* 1912. July 29. Vol. 49. No. 31. pp. 1453-1455.

In a previous paper the authors have shown that bodies of the safranine group possess a specific trypanocidal action, and Krause working in conjunction with Weber has adduced evidence that this action depends upon the side chains in the molecule, the removal from or the addition to the side chains of a group causing the drug to become increasingly or decreasingly active.

The authors state that the failure of the drugs employed by other authors to effect a radical cure is probably due to the fact that they act on the parasites present in the blood only and are too rapidly split up, the trypanosomes in the organs and tissues escaping their action. Their object therefore was to discover some substances which were stable and which would be able to reach all tissues. They claim to have found such substances belonging to the Safranin and Eurhodin groups.

The characteristic features of these dyes are that they contain in their molecules a combination of trivalent and pentavalent nitrogen and that they possess an extraordinarily active trypanocidal action, even when administered by the mouth. They are quite stable and in the pure state non-toxic. Thirty or more grammes may be given to the human subject in doses of $\frac{1}{4}$ to 1 gramme daily without any secondary effects. The substances contain no arsenic.

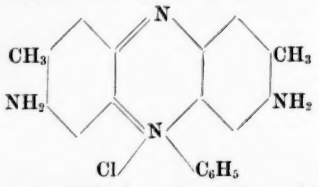
The animals used for the experiments were rats and guinea-pigs and four strains of *T. brucei* were employed. The results were the same with each of the strains. Series of infected animals were fed with the drugs, some being kept as controls in every case. All the animals receiving the substances were cured, that is to say they remained free from trypanosomes for months. The drugs were mixed with moistened bread for the rats and with moist bran for the guinea-pigs, and the dose was from 0·05 to 0·1 gramme. The feeding was continued for 4-6 weeks and every other day food was given with the admixture of the dyes.

Rats which had been free from trypanosomes for months were more difficult to infect than fresh animals, and the infection in such rats was more easily overcome. It was found that all compounds were not equally valuable. Some compounds which were actively trypanocidal entirely lost that power when their molecule was a little altered. With the most suitable compounds the

authors were able to free guinea-pigs from trypanosomes six days after inoculation and twenty-four hours before death, and to produce an absolute cure with further doses. The control guinea-pigs in the same experiments died in about seventeen days.

The sera of uninfected animals which were given the drug daily for some time exercised no protective or curative effect.

A substance having the following composition was very active:



Apparently the symmetrical arrangement of the methyl and the amido groups is connected in some way with the action. If one of the methyl groups be removed not only is the colour of the substance changed, but the trypanocidal action is nearly destroyed.

If two methyl groups are introduced into one of the amido groups serious sequelae follow the administration of the compound. The removal of both methyl groups restores the trypanocidal action, but the substance so obtained is not so active as the first compound mentioned.

The effect of methyl and amido groups on the trypanocidal action of drugs has already been shown by Krause in the dyes of the triphenylmethane group.

A number of other details are given by the authors regarding the effects produced by modifying the side chains of the molecule. In some cases there is loss of trypanocidal effect, in others toxic effect are produced, and so on.

Other bodies of the safranine group, such as indulin-scharlach, were tried, but their trypanocidal action was weak, as also was the action of the eurhodin group.

(129) Castelli (G.). **Chemotherapeutische Versuche über die Wirkung des Kakodyl und Arrhenal bei experimentellen Spirillen- und Trypanosomenerkrankungen.** [Experiments with Kakodyl and Arrhenal in Experimental Spirillosis and Trypanosomiasis.]—*Arch. f. Schiffs- und Trop. Hyg.* 1912. Sept. Vol. 16. No. 18. pp. 605-619.

The author's experiments had for their object the determination of the toxicity of these two compounds for the mouse, fowl and rabbit, and their curative effect on recurrent fever, trypanosomiasis (nagana), spirochaetosis of the fowl, experimental syphilis and yaws.

It was found that the maximum safe dose (dosis tolerata) of sodium kakodylate for the mouse by subcutaneous injection was 0·02 g. per 20 g. body weight. For the fowl intramuscularly the dose was 0·6 per kilo, and for the rabbit by intravenous injection 0·3 g. per kilo.

The following results were arrived at in experiments with arrhenal. Maximum safe dose for the mouse subcutaneously

0·5 g. per 20 g. body weight; 1·6 g. per kilo. for the fowl intramuscularly; and 0·5 g. intravenously for the rabbit.

Experiments to test the curative value of the compounds in the diseases mentioned showed that they were absolutely without effect.

(130) Laveran (A.) & Roudsky (D.). **Au Sujet de l'Action de l'Akridine (Diphénylméthane) sur** *Trypanosoma lewisi* **et** *Trypanosoma duttoni.* [The Action of Acridine on *T. lewisi* and *T. duttoni.*]—*Compt. Rend. Soc. Biol.* 1912. July 26. Vol. 73. No. 27. pp. 172-175.

In a previous publication the authors have shown that oxazine and acridine have a remarkable affinity for the substance of the centrosome of trypanosomes and they have come to the conclusion that the destruction of the centrosome is in all probability due to a process of auto-oxidation.

According to Kudicke the centrosome of *T. lewisi* in rats treated by acridine disappears in a definite fashion. According to this author one porton of the centrosome becomes displaced towards the anterior portion of the body while the origin of the flagellum undergoes no alteration. The portion which alters its position is reabsorbed or eliminated *in toto*. Kudicke has never observed the penetration into or the fusion of this portion of the centrosome with the nucleus. If acridine is administered during the acute stage when multiplication forms of the parasite are numerous the process according to Kudicke is different. At the moment when certain trypanosomes are undergoing division the centrosome itself does not divide, one of the daughter trypanosomes thus being left without a centrosome. This abnormal division which has already been described by Werbitzki would be the starting point of the acentrosomic varieties of the trypanosome.

Up to the present the authors have not observed the changes described by Kudicke, the explanation being that they have only studied the action of oxazine on pathogenic trypanosomes, and the peculiar phenomenon observed by Kudicke apparently occurs only in animals infected with non-pathogenic trypanosomes and treated by acridine. A more recent series of experiments carried out with acridine on rats infected with *T. duttoni* has confirmed Kudicke's results. A rat inoculated with *T. duttoni* showed numerous trypanosomes in its blood on the fourth day after inoculation and it was given a quarter of a milligramme of acridine in a freshly prepared solution. On the following day a certain number of the trypanosomes were observed to possess no centrosome while in others the centrosome was free in the cytoplasm. The position of the free centrosome was variable. The posterior extremity of the flagellum was not altered in position but in a number of cases showed a slight thickening. Two days later the rat received a second injection of acridine, the same dose being administered. On the following day the number of acentrosomic trypanosomes was increased and the rat was killed for the purpose of inoculating a fresh one. It appears to be probable that under the action of acridine the centrosome becomes more friable and as a result of the movements of the flagellum becomes detached and is thus left

free in the cytoplasm. The authors have examined trypanosomes with free centrosomes coloured by oxazine or acridine *in vitro*, and they have been able to determine that the movements of the free centrosomes are purely passive. These centrosomes are gradually absorbed thus producing acentrosomic trypanosomes. Before disappearance is complete the centrosome is in some cases reduced to a collection of fine granules which can be stained *in vitro* by acridine or oxazine. The absence of such effects of acridine upon the centrosomes of pathogenic trypanosomes may be explained as follows:—These centrosomes which are much smaller than those of *T. lewisi* and *T. duttoni* offer less resistance to the traction exerted upon them by the flagellum and consequently are less likely to become detached. Acridine also has far less effect upon the centrosomes of the pathogenic trypanosomes than upon those of *T. lewisi* and *T. duttoni*. On the other hand oxazine has far less action on the latter than upon the majority of the pathogenic trypanosomes.

(131) Pecaud (G.). **Contribution au Traitement des Trypanosomaises Animales.** [The Treatment of Animal Trypanosomiases.]—*Bull. Soc. Path. Exot.* 1912. June. Vol. 5. No. 6. pp. 385-389.

Cattle appear to be more susceptible to orpiment than horses, and the maximum dose should not exceed 7-8 grammes per 100 kilo. live weight. The administration of the drug is difficult in cattle. If given in the form of a ball there is a risk of the ball resting in the rumen or abomasum and setting up severe inflammatory conditions which may lead to gastric fistula and necrosis of the stomach and skin. For this reason the author administered the orpiment in the form of an electuary.

In dealing with cattle the following plan was resorted to: The electuary was given every two days, the dose to commence with being about 4 grammes per 100 kilog. The dose was gradually increased until the maximum dose was given at the fourth or fifth administration. This last dose was repeated once.

As a general rule the trypanosomes disappeared from the blood after the first dose, and this was particularly the case with *T. cazalboui*.

In some instances they reappeared eight to ten days after the first series of doses, in which case the treatment was repeated.

The mixed method of treatment involves the alternate administration of orpiment and atoxyl, the latter being given subcutaneously in doses of 2 to 3 g. at a time.

The author prefers to use orpiment alone as it is just as effective and less costly, but orpiment may be replaced by an injection of atoxyl when the animal shows symptoms of poisoning.

In any case no definite rules can be laid down, cases must be treated on their merits. Great care has to be taken in administering orpiment because there is very little difference between the toxic and the therapeutic dose, and it is of the utmost importance to use the pure precipitated drug.

Relapses are not uncommon, and in such cases treatment has to be commenced over again.

The author has obtained similar results with tartar emetic, and he finds that it is easier to administer. He follows the plan already advised by BRODEN and RHODAIN, and administers the drug subcutaneously, using a 2 per cent. solution for the purpose. In the majority of cases four or five administrations of 1 to 2 g. suffice.

It would appear that orpiment is more effective against *T. cazalboui*, while emetic is more effective with *T. dimorphon*.

Neither drug appears to have any permanent effect on *T. pecaudi* in the horse.

The following are the results obtained and in every case the animals have been followed for a period of two years or more.

Cattle.—T. dimorphon.—Emetic. Eight cases treated, six recoveries and two deaths.

Orpiment alone.—Twenty-four cases treated, eighteen recoveries, three deaths, and three cases of gastric fistula as a result of administering the drug *in bolus*.

Orpiment-atoxyl.—Seven cases treated, one recovery, four deaths, and two deaths from gastric fistula.

Horses.—T. pecaudi.—Atoxyl-orpiment.—Three cases treated, all failures.

T. cazalboui.—Five cases treated, one death, two successes, one relapse successfully treated, and one relapse which was not treated and proved fatal.

Donkeys.—T. cazalboui.—Two cases treated. One death, and one success, but the animal aborted.

Comparing the mortality among treated and untreated animals the author finds that while among untreated animals the mortality has been 90 per cent. and none of the pregnant females have had a normal parturition, in treated animals (excluding accidents) the mortality has been 26 per cent., and abortion has occurred in 37·5 per cent. of cases.

The author adds that in every case the animals put under treatment had been affected for a short time only.

(132) MATTES (W.). **Agglutinationserscheinungen bei den Trypanosomen der Schlafkrankheit, Nagana, Dourine, Beschalseuche und des Kongokustenfiebers, unter Berucksichtigung der Farbemethoden, der Morphologischen und biologischen Verhaltnisse der Erreger.** [Agglutination of the Trypanosomes of Sleeping Sickness, Nagana, Dourine, and Congo Coast Fever, and Consideration of the Staining Characters and Morphological and Biological Characters of the Organisms.]—*Centralb. f. Bakt.* 1. Abt., Orig. 1912. Aug. 10. Vol. 65. Nos. 6-7. pp. 538-573.

The procedures adopted and the results obtained by a number of other observers are briefly referred to and a somewhat extensive description of the morphology, developmental forms, pathogenicity, vitality, cultivation, symptoms, lesions, and staining characters of the various species of trypanosomes used are given before the author proceeds to the description of his own experiments in connection with the agglutination of trypanosomes.

The following species of trypanosomes were used in the experiments: *T. gambiense, T. brucei, T. equiperdum, T. congolense,*

and the trypanosome of dourine. According to the author *T. equiperdum* is the cause of dourine (*Beschälseuche*) which occurs extensively in Europe, and the parasite referred to as the trypanosome of dourine is the organism responsible for the disease in Algeria.

The author points out the differences between the agglutination test as carried out with bacteria and with trypanosomes, emphasising the fact that trypanosomes for the purpose must be obtained from an infected animal and also the supreme importance of absolute sterility of all the instruments and vessels.

In the majoriy of cases the author obtained his suspension of trypanosomes by bleeding animals at the height of infection into citrate solution. He found it inadvisable to chloroform the animal, because this lessened the amount of blood obtainable, and he also feared that the chloroform might have some effect upon the agglutination. The mixture thus obtained was centrifuged in the water centrifuge because it was observed that if the electric centrifuge were used at 4,000 revolutions per minute the trypanosomes were so clumped together as to be useless. Following LANGE's procedure the author found it difficult to obtain a suspension of trypanosomes free from red blood corpuscles, he therefore devised the following methods. The tubes used for centrifuging the diluted blood were about 5 mm. in diameter, and the lower end was drawn out into a capilliary tube about 10 cm. long and sealed up. During centrifugalisation the tubes were carefully packed round with wool to prevent breakage. After complete sedimentation of the corpuscles the capillary tube was broken at the junction of the sedimented corpuscles and the layer of liquid containing the trypanosomes, the upper end of the tube being closed to prevent the liquid from flowing out. The trypanosomes thus obtained were thrice washed.

The second plan devised by the author was to stop the centrifuging at the moment when the great majority of the red cells had been thrown down, but the trypanosomes were still to a great extent in suspension. The supernatant liquid containing the trypanosomes and a few red corpuscles was drawn off and again centrifuged, the process being continued till sedimentation was complete. A delicate capillary pipette was then introduced and the trypanosomes stirred up into the liquid by careful rotation of the point of the pipette around the tube just above the layer of trypanosomes. Proceeding in this way, and if necessary adding more salt solution and re-centrifuging, the majority of the trypanosomes could be obtained.

The suspension so obtained had then added to it five to ten per cent. of 15 per cent. formalin in salt solution. Experiments were made without success to replace the formalin by carbolic acid, cresol, chloroform, and absolute alcohol.

About two to three cubic centimetres of suspension are to be obtained from a large mouse.

Spontaneous agglutination of trypanosomes in the suspension is likely to occur if too large a proportion of formalin be added, and also if the suspension be allowed to stand before the formalin is added. The sera used in the tests were obtained from man, the

horse, ox, sheep, goat, pig, rabbit, guinea-pig, and mouse, and in no case was any preservative added.

The mixtures of serum and suspension were kept sterile by means of plugs during the test, and the tubes were incubated for twelve or more hours at 37° C. In some cases it was necessary to incubate for thirty to forty hours before any reaction became evident. The author found that normal sera were capable of agglutinating trypanosomes in much higher dilutions than normal sera generally agglutinate bacteria. Tests were made with each of the species of trypanosome using normal sera from the animals mentioned, but in no case was any agglutination observed in dilutions higher than 1 in 100. The sera of the horse and the ox were found to possess the highest power, and in practically no other case was any agglutination observed in dilutions higher than 1 in 10.

Agglutinating power of immune sera. In order to obtain as large an amount of agglutinin as possible the author first inoculated his animals with either dead trypanosomes, or trypanosomes that had lost a great deal of their vitality through the blood being allowed to stand after withdrawal for a time, and then again inoculating with living fully virulent trypanosomes after an interval of about a week. In the case of the least pathogenic trypanosome —the *congolense*—a third inoculation was given. Blood was withdrawn at intervals and the agglutinating power tested, and the animals were killed when seriously ill. It was found that there was a somewhat rapid elevation in the amount of agglutinin present.

Tests were made at intervals with homologous and heterologous trypanosomes. From these it was found that the reactions are not specific, a nagana-immune serum agglutinates not only *T. brucei* but all the other species used in the tests, but the agglutination with the homologous strain occurs in higher dilutions than with the heterologous strains. For example, the serum of a rabbit inoculated with nagana agglutinated the *T. brucei* in a dilution of 1 in 3,000, the dourine trypanosomes in dilutions of 1 in 2,000, and the *T. congolense* and *T. gambiense* in dilutions of 1 in 1,500.

The author found that agglutinin is present in exudates in the serous cavities as well as in the blood.

The sera of two horses that had recovered from dourine were tested with suspensions of the different trypanosomes and similar results were obtained.

The results were not influenced by inactivating the serum by heating for some hours at 60° C., nor did heating the suspension have any effect on the results. Exactly similar results were also obtained when the suspension was cooled to — 25° C. for three hours.

A number of experiments were made in which the serum was diluted and the suspension made with salt solution that had been rendered radio-active. In two instances there was spontaneous agglutination in suspensions so prepared without the addition of any serum. The experiments made with sera lead the author to conclude that the agglutination occurred in much higher dilutions and more rapidly when radio-active salt solution was used than when ordinary salt solution was employed.

(133) Schilling (C.). **Ein neues Immunisierungsverfahren gegen Trypanosomen-Infektionen.** [A New Method of Immunising against Trypanosome Diseases.]—*Deut. Med. Wochenschr.* 1912. Aug. 22. Vol. 38. No. 34. pp. 1579-1580.

The principle upon which the method is based is the inoculation of susceptible animals with trypanosomes which have been killed by chemical substances, but which, according to the author, still retain their power of provoking the formation of antibodies.

Experiments have been made with two horses using the *T. brucei*. It would appear that the trypanosomes are obtained free from blood and treated for two hours with an equal quantity of 1 : 400 solution of tartar emetic. This liquid is centrifuged and the sediment used as the antigen.

The first horse treated received seven injections, some subcutaneous and some intraperitoneal, of trypanosomes treated in this way obtained from a total of fifty infected rats, the injections being spread over a period of ten weeks. A week after the last injection the horse received an intravenous inoculation of 0·5 cc. of mouse blood containing *T. brucei*. The horse failed to become infected.

The second horse was treated in a similar manner, but only five injections of dead trypanosomes were given. When tested with 0·5 cc. of mouse blood containing trypanosomes the animal failed to become infected.

No definite results have been obtained in experiments in which increasing quantities of serum from treated animals were mixed with trypanosomes and inoculated into the peritoneal cavity of mice, with the object of standardising the serum.

The duration of the immunity was tested by reinoculating a number of mice at intervals after they had been protected and inoculated once. The results were inconstant. Whereas two mice were reinfected after an interval of two months, two others resisted this inoculation and two further inoculations at intervals of one and two months.

(134) Duke (H. L.). **Antelope as a Reservoir for *Trypanosoma gambiense*.**—*Proc. Roy. Soc.* 1912. July 25. Series B. Vol. 85. No. B. 579. pp. 299-311.

In considering the problem of the prolonged infectivity of *G. palpalis* on the shore of Lake Victoria Nyanza the subject must be approached from three points of view, (1) some idea of the percentage of infected wild flies must be obtained from time to time in order to gauge the efficiency of the reservoir, (2) the wild animals frequenting the Lake shore must be examined for *T. gambiense*, and (3) the effect of *T. gambiense* on these animals should be studied in the laboratory.

It has been pointed out in the Report covering the period March, 1911, to January, 1912, that the percentage of infected flies along the Chagwe coast line is 0·014 per cent. During the interim no natives have been allowed in the neighbourhood of the fly except those in the employ of the laboratory.

On Damba Island in May, 1911, Carpenter obtained infection of a monkey with *T. gambiense* using 880 wild fly, and two out

of four animals shot on this island were found to harbour the trypanosome. The conditions on this island are exceptional in that the fly and antelope are frequently brought into contact.

Under such conditions it would appear necessary for the antelope to remain infected over a very long period in order to keep up the infectivity of the fly, and nothing short of a very rapidly acquired immunity to the disease on the part of the antelope could prevent a relatively large number of fly being infective at any given time. On the main land the opportunities for the fly to bite these animals are much less frequent, and, bearing in mind the small percentage of flies capable of carrying the trypanosome, it becomes of great importance to know how long an antelope can remain infective.

It has been shown that these animals may remain infective to fly for a period of at least 10 months. Up to May, 1911, the evidence of infectivity afforded by the presence of trypanosomes in the laboratory bred *G. palpalis* fed upon the buck proved sufficiently regular without resorting to blood injection. The experiments given in this paper represent the continuation of these investigations up to January, 1912. During this period laboratory bred *G. palpalis* were fed upon the antelope and their blood was also tested by injection into susceptible animals. The injection test gradually came to supersede the employment of clean flies.

Details are given of fly experiments and injection experiments with six antelope, and the longest period for which one of these was found to remain infective was 22 months after its original infection with laboratory bred *G. palpalis*.

Brief details are also given regarding experiments which were undertaken to ascertain whether the serum of these antelope had acquired any immunising properties. The author concludes that there is some evidence to show that an antelope which has ceased to be infective for *T. gambiense* acquires some degree of immunity against re-infection.

(135) Kinghorn (Allan) & Yorke (Warrington). **On the Transmission of Human Trypanosomes by *Glossina morsitans*, Westw.; and on the Occurrence of Human Trypanosomes in Game.**—*Ann. Trop. Med. & Parasit.* 1912. March 29. Vol. 6. No. 1. A. pp. 1-23.

The authors' summary is as follows:—

"1. The human trypanosome, in the Luangwa Valley, is transmitted by *Glossina morsitans*, Westw.

"2. Approximately 5 per cent. (4·76) of the flies may become permanently infected, and capable of transmitting the virus.

"3. The period which elapses between the infecting feed of the flies and the date on which they become infective, is approximately fourteen days.

"4. An infected fly retains the power of transmitting the disease during its life, and is infective at each meal.

"5. Mechanical transmission does not occur if a period of twenty-four hours has elapsed since the infecting meal.

"6. Some evidence exists to shew that in the interval between the infecting feed and the date on which transmission is possible, the parasites found in the flies are non-effective.

"7. *Glossina morsitans*, in nature, has been found to transmit the human trypanosome.

"8. Certain species of buck, viz., waterbuck, hartebeest, mpala, and warthog, have been found to be infected with the human trypanosome.

"9. A native dog has been found to be infected with the human trypanosome."

(136) Duke (H. L.). **Observations on Fowls and Ducks in Uganda with Relation to *Trypanosoma gallinarum* and *T. gambiense*.** (With a Note by Miss Muriel Robertson.)—*Proc. Roy. Soc.* 1912. August 20. Series B. Vol. 85. No. B. 580. pp. 378-384.

In the course of a considerable number of experiments carried out to test the duration of infectivity of antelopes for *T. gambiense* Uganda fowls were employed to nourish the flies during the earlier days of the experiment. In the course of routine dissections of laboratory bred flies from these experiments a curious crithidial type of flagellate was noticed in the hinder part of the mid-gut. In morphology and movement the organisms were immediately distinguishable from the developmental stages of the *T. gambiense* in *G. palpalis*. As some of the laboratory antelope had been found to be infected with *T. ingens* it appeared possible that the flagellates might represent developmental stages of this trypanosome. On the other hand suspicion fell upon *T. gallinarum*, although microscopic examination of the fowls used had always proved negative. The connection between *T. gallinarum* and these crithidiae was established by finding them in clean laboratory bred flies fed upon the suspected cocks and by the subsequent discovery of the trypanosomes in the cocks' blood. The trypanosomes are very rare in the peripheral circulation.

The following is a summary of Miss Robertson's note on the morphology of the parasites:—In many respects the appearance of *T. gallinarum* in *G. palpalis* is exceedingly characteristic. The trypanosomes are, as a rule, situated in the middle and posterior portions of the mid-gut. The motion of the flagellate in the live state is very striking, the body in the vast majority of cases is quite stiff through about two-thirds of its length and is often broad and relatively massive. The flagellum is much thicker than in mammalian trypanosomes. Progression takes place in straight lines, and in the more slender individuals may occasionally be reversed in direction. The breadth of the body is subject to variation, and as it is impossible to tell when the flies first receive the trypanosomes it is impossible to give the exact sequence of the forms in the cycle. Apparently, however, the massive broad forms gradually give place to smaller and more slender individuals, some of which show considerable lengthening of the body.

True trypanosome phases have not been observed. Although binary fission appears to be the usual method of multiplication, multiple division is not uncommonly observed.

Morphology.—Films were fixed wet with Schaudinn's fluid and stained with iron-haematoxylin and haemalum.

The protoplasm especially in the broader forms is very dense and granular. The tropho-nucleus is of a common trypanosome

type, consisting of a large central karyosome surrounded by a clear space which is bounded in turn by a delicate membrane. Fine rays pass from the karyosome to the membrane.

The kineto-nucleus lies close to and generally in front of the tropho-nucleus. It is relatively large and presents the somewhat curious double appearance of two closely-apposed granules lying one behind the other. This does not seem to be in any way a case of precocious division. The blepharoplast proper, *i.e.*, the minute granule actually at the origin of the flagellum is usually obscure but can be seen in favourable specimens.

Another structure not usually observed in trypanosomes is a granule situated towards the posterior end of the cell. The outline of this granule is rarely very sharply defined and in some instances it appears to be attached by a fine line to the nucleus or to the blepharoplast. The granule divides when the parasite divides.

A few experiments were undertaken to ascertain whether ducks were capable of acting as a reservoir for *T. gambiense* and whether newly arrived ducks were capable of infecting laboratory bred flies with flagellates. The results in both cases were negative.

(137) Mesnil (F.) & Blanchard (M.). **Infection des Poules dues aux *Trypanosoma gambiense* et *Trypanosoma rhodesiense*.** [Infection of Fowls with *T. gambiense* & *T. rhodesiense*.]—*Compt. Rend. Soc. Biol.* 1912. June 14. Vol. 72. No. 21. pp. 938-940.

The authors inoculated four fowls each with *T. gambiense* and *T. rhodesiense*. The inoculations were successful in every instance in the former and in three of the birds in the latter. Trypanosomes were never found in moist preparations of blood, but the blood was proved to be infective by inoculation. In two birds a few trypanosomes were seen in the leucocyte layer of blood taken at the time of death and centrifuged. Three of the fowls inoculated with *T. gambiense* died in one to three months. In the fourth bird the disease lasted for a month and rats subsequently inoculated failed to become infected, and the bird was thought to have recovered. It was reinoculated and died ten days later, its blood proving infective for mice.

Two of the birds infected with *T. rhodesiense* died in about two months.

The blood of the third was proved to be infective up to the fifty-fifth day, and the bird died on the eighty-third day, its blood being then non-infective. Death was due to pasteurellosis. The fourth fowl died of pasteurellosis without becoming infected.

There were lesions involving the eyes of the birds that died of the infection in both cases.

The serum of the birds infected with *T. rhodesiense* mixed with the homologous trypanosome retarded the evolution of the disease in a mouse for three or four days, but had no action on the heterologous trypanosome. The serum of birds infected with *T. gambiense* had no effect upon either.

(138) Bayon (H.). **The Cultivation of *Trypanosoma rhodesiense*, Stephens and Fantham.**—*Proc. Roy. Soc.* 1912. August 24. Series B. Vol. 85. No. B. 581. pp. 482-483.

The author has been able to cultivate *T. rhodesiense* successfully on the following media, (1) Clegg's *Amoeba* agar to which is added twice its amount of rabbit's blood which has been frozen and thawed rapidly so as to cause the haemoglobin to diffuse into the serum; and (2) the following formula:—agar 15 grammes, glucose 10 grammes, water 1,000 grammes, and twice the volume of rabbit's blood.

The blood containing the trypanosomes was placed on the agar which forms a diffluent mass at the bottom of the tube. The tubes were incubated at 22° to 25° C. Multiplication could be observed under the microscope, and the cultivated trypanosomes remained actively motile up to the twenty-first day of cultivation. They were virulent up to the eighth day of culture. In certain cases rosette forms were observed and true-brood forms have also been distinguished. In some of the tubes rounded forms appeared on the third to the fifth day; these are being separately studied. The percentage of successes varied from one tube in twenty-five to six in six.

(139) Yorke (Warrington) & Blacklock (B.). **A Note on the Morphology of a Strain of *Trypanosoma equiperdum*.**—*Brit. Med. Jl.* 1912. August 31. p. 473. With 1 chart and 13 text-figures.

The occurrence of posterior nuclear forms of this parasite had already been noticed by the authors in smears of guinea-pig blood infected with this trypanosome some months before the present observations were made.

More recently the authors obtained a fresh strain of dourine virus from Schilling, the strain having been running in his laboratory for many years.

Morphology. Between the long thin forms with free flagellum and short stumpy forms without free flagellum there is a variety of intermediate forms.

A thousand parasites from a white rat, drawn and measured over a period of eight days before its death, had an average length of 26·7 microns. The longest measured 36 and the shortest 15 microns.

The forms of the parasite in which the nucleus is observed to lie posterior to the centre are the short stumpy types which have little or no free flagellum. They have been observed in rabbits, guinea-pigs and rats—the only animals that have been experimented with. The nucleus varies in position from being just posterior to the centre of the body to close to the blepharoplast. A definite posterior position of the nucleus was found in 3·8 per cent. of a thousand trypanosomes. Many of the parasites have very marked granules.

(140) Wenyon (C. M.). **The Insufficiency of the Posterior Nucleus as a Specific Distinction in** *Trypanosoma rhodesiense*.—*Jl. Trop. Med. & Hyg.* 1912. July 1. Vol. 15. No. 13. pp. 193.

The figures of *T. rhodesiense* given by Stephens and Fantham reminded the author of *T. pecaudi*, and on looking through some films of rat blood containing *T. pecaudi* which he made in the Sudan in 1908 he found that forms with the trophonucleus near to the kinetonucleus are fairly common, while a small percentage of the comparatively broad trypanosomes have the nucleus at the posterior end of the body.

In the light of the diagnostic importance of this arrangement of the nucleus in *T. rhodesiense*, as emphasised by Stephens and Fantham, the author thought it worth while to place on record the present observations.

In the case of *T. pecaudi*, the forms with the nucleus near the posterior end occurred both in the blood of inoculated rats and in the blood of the original host—a donkey. In the latter the author has not found a specimen of the trypanosome with the nucleus on the posterior side of the kinetonucleus, but thinks it probable that prolonged search would reveal such, since in many the nucleus is right up against the kinetonucleus. In the case of *T. rhodesiense* the posterior nuclear forms occur in sub-inoculated rats only, whereas in *T. pecaudi* they occur in the donkey also. This is probably dependent upon the infection, which is always very small in man, while it was heavy in the donkey. In the case of both these trypanosomes it is perhaps incorrect to speak of either man or donkey as the true host. In neither case is there any sign of adaptation, for the infection nearly always brings about a fatal termination. The true host is the reservoir from which man and the donkey become infected.

It is quite possible that one and the same trypanosome would show certain morphological variations in the same host, provided that in one district the host had become more adapted to the parasite than in the other. It would be interesting to know whether *T. pecaudi* shows posterior forms in the reservior host, but this has apparently not yet been discovered.

Drawings of nine trypanosomes are given and the author points out that the broad forms with the posterior nucleus divide and give rise to trypanosomes of the ordinary type.

(141) Roudsky (D.). **Action Pathogène de** *Trypanosoma duttoni* **Thiroux, et Lésions Provoquées chez le Rat par ce flagellé.** [The Pathogenic Effect of *Trypanosoma duttoni*, and the Lesions caused by the Organism in the Rat.]—*Compt. Rend. Soc. Biol.* 1912. July 26. Vol. 73. No. 27. pp. 170-172.

In a previous publication the author has shown that *T. duttoni* is capable of causing death in the rat. This pathogenic effect, which was practically constant in the first passages in series from rat to rat, gradually became attenuated as the number of passages increased. The author has made 53 passages and every rat has become infected, but death has been exceptional and haemoglobinuria very rare. The infection itself has become more brief since

the first passage. The pathogenic power acquired by *T. duttoni* by changing the vertebrate host is modified in a quite different manner to that of *T. lewisi* acclimatised for the mouse, the latter showing a progressive increase of virulence.

During the first passages from rat to rat *T. duttoni* showed characters different from those observed in the mouse. The posterior extremity became elongated to the extent of exceeding in some individuals the length of the anterior part. This posterior extremity was composed either of periplasm alone or of two layers of periplasm separated by a small amount of cytoplasm. In certain instances the elongated posterior extremity resembled a second flagellum in appearance. This morphological character suddenly disappeared at the 9th passage, the parasite losing its pathogenic power at the same time.

The centrosome in practically every parasite appeared bi-lobed.

Lesions were to be found in a number of the viscera of rats which died showing symptoms of haemoglobinuria as a result of infection with *T. duttoni*.

The hepatic cells showed a slight amount of coagulative necrosis around the central veins resulting from pressure by the engorged capillaries. Cellular infiltrations, consisting principally of mononuclear leucocytes, were found around the portal spaces. In one instance the author found giant cells.

In the kidneys a considerable number of the tubes were filled with an amorphous material. In some portions of the tubes there was complete destruction of the epithelium, the lumen being filled with a finely granular material which contained a number of desquamated cells more or less altered in appearance.

The parenchyma of the spleen was infiltrated with a large quantity of blood.

To the naked-eye reddish areas could be observed in the parenchyma of the lungs, the acini in these places being distended with an oedematous fluid.

The author concludes that the lesions produced may be due to a toxin developed by *T. duttoni* to which the rat is susceptible.

(142) Schepilewsky (E.). **Fadenförmige Anhängsel bei Trypanosomen.** [Thread-like Appendages in Trypanosomes.]—*Centralbl. f. Bakt.* 1. Abt. Orig. 1912. July 3. Vol. 65. Nos. 1-3. pp. 79-83. With 1 plate.

After a brief reference to the morphological characters of the *T. brucei* and *T. equiperdum*, the author passes to a consideration of the work of Wasielewski and Senn, who first observed marked elongation of the posterior extremity of trypanosomes in rat blood, and who considered the elongation to be an artefact due to the spreading of the blood. Wendelstadt and Fellmer who also observed this elongation did not agree that it should be referred to an artefact because it was not observed to be all in one direction. The work of v. Prowazek, Doflein, Leporsky, Yamamoto and others, is mentioned.

The author's own observations have been made with dark-ground illumination, and the two trypanosomes used for the

observations were those mentioned above, the strains having been kept in guinea-pigs and mice. The blood used for examination was mixed with salt solution in such proportions that under the microscope there was a certain amount of space between the blood corpuscles. The preparations had to be examined immediately they were made as the trypanosomes died in about half an hour.

With dark ground illumination it can be quite clearly made out that the bodies of the parasites are surrounded by a thin refractile envelope which is sharply defined both externally and where it abuts on the cytoplasm. This envelope (pellicle) extends to the anterior end of the flagellum giving it a double-contoured appearance. The double contour seems to disappear at the extreme end of the flagellum under a comparatively low magnification, but it can be easily made out under the oil immersion. For this reason it cannot be asserted that the two trypanosomes under consideration have in reality any free flagellum.

No structure can be made out in the endoplasm with dark-ground illumination while the trypanosome remains alive, and the nucleus is not visible. Sometimes one or more refractile particles can be made out in the cytoplasm. Immediately the trypanosomes die the whole of the protoplasm becomes granular, and the nucleus appears as a dark spot.

The author has been able to observe details which do not appear to have been described before. In the living parasites examined with the dark-ground illumination he has been able to make out thread-like appendages attached either to the end of the flagellum, or to the posterior end of the body, or both. These appendages are not always present. He was never able to observe them attached laterally to the trypanosomes. The threads are very delicate. They are of constant thickness throughout and are only slightly refractile. The point where the flagellum and the thread-like appendage meet is clearly visible owing to the difference in thickness and the difference in refractility. The threads vary in length, but as the rule the anterior ones are longer than the posterior, and may be as much as twice the length of the body. Occasionally the author observed double or divided threads. The posterior threads as a rule remain extended, but the anterior ones move with the flagellum. In some cases the end of the thread remains attached to the cover-glass and thus fixes the trypanosome to a certain extent.

Apparently the threads possess no power of motion of their own. They are very easily destroyed. In a dying trypanosome they disappear in a few minutes. They first become pale, then granular, and, finally, disappear as if dissolved.

These structures cannot be confounded with those described by Wasielewski and Senn, or Wendelstadt and Fellmer, because the bodies seen by those authors occurred at the posterior end of the body only and were observed in stained preparations. Nor can they be confused with the free flagella described by v. Prowazek because they are not connected with the blepharoplast.

The author, while unable to form any opinion as to the significance of these appendages, believes that they are composed of the protoplasmic substance.

(143) Nattan-Larrier (L.). **Non-Transmission des Trypanosomiases de la Mère au Foetus.** [The Non-Transmission of Trypanosomiases from the Mother to the Foetus.]—*Bull. Soc. Path. Exot.* 1912. Vol. 5. No. 7. pp. 550-556.

Difficulties were encountered in the experiments made in connection with this question owing to the frequency with which abortion followed the inoculation of the females.

Two series of experiments were carried out simultaneously as follows:—

(1) Experiments to test whether trypanosomes are able to pass the placenta and gain access to the blood of the foetus.

(2) Experiments to ascertain whether the serum of the foetus acquires any special characters resulting from the infection of the mother.

(1) Twelve experiments in all were made in this connection, eight with guinea-pigs, two with rats, and two with mice.

The following was the technique employed in every case.

If parturition occurred spontaneously the young were immediately killed and their blood used for the inoculation of mice. In the majority of cases, however, the animals were killed before parturition occurred, the abdomen opened, and the vagina and fallopian tubes ligatured. The surface of the uterus was then extensively cauterised, an opening being made with a needle heated to redness. The amniotic fluid was withdrawn with a pipette, the opening in the uterus enlarged, the foetuses withdrawn, and the umbilical cords ligatured and cut.

The foetuses were washed in sublimate and salt solution, the blood being withdrawn from the heart with a pipette. In every case about a dozen drops of amniotic fluid and a similar quantity of blood were used for the inoculation of a number of mice. Finally both the blood and the amniotic fluid were examined microscopically.

Proceeding in this way sixteen mice were inoculated, but the result in every case was negative. The trypanosomes used were: *T. evansi*, *T. brucei*, *T. congolense*, *T. soudanese*, and *T. gambiense*.

The author thinks that before expressing an opinion as to whether this holds true for all species of trypanosomes experiments should be made with *T. equiperdum* since this organism is able to pass through intact mucous membranes. He also suggests that *Schizotrypanum cruzi* might also be capable of causing hereditary lesions.

2. Experiments to ascertain whether the serum of the progeny of infected females acquires any special characters.

In this connection two points have been investigated, viz., whether the serum possesses any protective power, and whether it is capable of causing agglutination.

Five experiments were made to test the serum of the young animals as to the presence of protective substances, the results being controlled by parallel tests with the serum of the mother. In one case both sera exercised a complete protective effect, in two cases the two sera produced a delay in the development

of infection to almost the same degree, and in the other two cases there was no effect whatever. The activity of the amniotic fluid was always found to be inferior to that of the serum.

The author has completed a single experiment only regarding the agglutinating power of the sera, and the result shows that the two sera produce agglutination to practically the same extent.

(144) Wolbach (S. B.) & Binger (C. A. L.). **A Contribution to the Parasitology of Trypanosomiasis.**—*Jl. of Medical Research.* 1912. Sept. Vol. 27. No. 1. pp. 83-107. With 6 plates.

The purpose of the work upon which the paper was based was to study the distribution of the trypanosomes in the tissues, and to ascertain the factors concerned in the production of the lesions of trypanosomiasis. This was stimulated by the acquisition of a reliable method of staining trypanosomes in the tissues. The method is as follows:—

1. The pieces of tissue for examination must be taken immediately after the death of the infected animal. They should be cut into slices not more than two millimetres thick and fixed for at least forty-eight hours in a mixture consisting of two parts of saturated solution of corrosive sublimate in distilled water and one part of absolute alcohol. The pieces may remain in this mixture for weeks without harm.

2. The pieces are dehydrated in alcohol, clarified in cedar oil, and embedded in paraffin. The sections should not be more than four microns in thickness, and it is better to have them half that thickness as the stain has poor penetrating power.

3. The paraffin is removed with xylol, followed by graded alcohols and water.

4. Treat with the following solution of iodine for fifteen to twenty minutes: Saturated solution of iodine in ninety-five per cent. alcohol three or four cubic centimetres; seventy per cent. alcohol, one hundred cubic centimetres.

5. Treat with eighty per cent. alcohol until the yellow colour is removed. Wash with distilled water and place in half per cent. aqueous solution of hyposulphite of sodium for ten minutes. Wash in running water for five minutes and rinse in distilled water.

6. Stain in freshly prepared Giemsa solution (60 to 80 drops to 100 c.c. of water) for four to twelve hours. The stain should be replaced twice by fresh mixtures during the first hour. The water used for diluting the stain must be absolutely free from acids.

7. Transfer the sections for differentiation through two changes of colophonium in acetone (15 per cent. solution). Fifteen to twenty seconds generally suffice.

8. Pass the sections rapidly through acetone-xylol (70 to 30), xylol, cedar oil, and mount in cedar oil.

According to the authors this method gives the same effect as that obtained in film preparations.

Two strains of trypanosomes were used during the investigation; a strain of *T. brucei*, originally brought from Uganda, and a strain of *T. gambiense* from the French Congo. Specimens for examination were obtained from four white rats, six guinea-pigs, and three monkeys infected with the *T. gambiense*, and from four white rats and three guinea-pigs infected with the *T. brucei*.

The authors state that they have not been able to make any substantial contribution to the pathology of trypanosomiasis, but they note the development of skin lesions associated with giant cell formation in the guinea-pigs infected with *T. gambiense*.

The following conclusions are drawn.

"In trypanosomiasis the trypanosomes do not remain confined to the blood vessels or the lymphatics. They invade the connective tissue structures of all the organs, the reticular tissue of the lymph nodes and spleen, and the substance of the brain.

"The lesions of trypanosomiasis are due to the presence of the trypanosomes in the tissues.

"The most common form of trypanosome in tissue, and we believe the one most active in the production of lesions, is the flagellate form."

SPIROCHAETOSIS.

(145) Deutz. **Ueber Versuche zur Uebertragung von Hühnerspirochaeten auf Mause.** [The Transmission of the Spirochaete of the Fowl to the Mouse.]—*Hyg. Rundschau.* 1912. Aug. 15. Vol. 22. No. 16. pp. 1017-1019.

The experiments of Levaditi and Lange have shown that the spirochaete of the fowl produces in rabbits inoculated with it a very benign disease only. The author therefore investigated the effect of inoculating mice with this organism. The mice used were inoculated either intravenously into the veins of the tail or intraperitoneally. The results varied in different cases. Whereas in some of the mice spirochaetes could be found in the blood 72 hours after inoculation, in others none could be found only 24 and 48 hours after. As the parasites disappeared from the circulation certain abnormalities of the blood made their appearance. There appeared to be an increase in the number of the blood corpuscles and blood platelets. The leucocytes appeared to be larger, and vacuole formation and amoeboid movements were observed to be far more prominent than normal. Appearances suggested that a process of phagocytosis was going on, and observations made at intervals of 10 to 15 minutes showed that such was the case.

Attempts were made to infect mice from each other by intravenous inoculation, but in no case was this successful for more than two passages. Using mice from 10 to 20 days old, three passages were obtained when the inoculations were intraperitoneal. As a general rule spirochaetes disappeared from the blood within 40 hours after intraperitoneal inoculation. Examination of the peritoneal fluid showed that within the course of the first hour after intraperitoneal inoculation a proportion of the organisms were ingested with phagocytes. Mice that have been inoculated with blood containing the organism fail to react to a second inoculation. If a recovered mouse be inoculated intraperitoneally, and the peritoneal fluid be examined immediately after the inoculation, typical agglomeration of the spirochaetes can be observed, the agglomeration being promptly followed by phagocytosis. The admixture of serum from a recovered mouse, or from one that has been repeatedly inoculated, with fresh actively motile spirochaetes causes an almost immediate cessation of movement. This is not observed if normal mouse serum be used. If the serum be taken from a mouse immediately spirochaetes have disappeared from its blood this phenomenon is not observed, the serum behaving exactly like normal serum.

Spirochaetes that have been passed through mice appear to have lost some of their virulence as indicated by the fact that when such spirochaetes are used to inoculate fowls the period elapsing between inoculation and death is about three times that observed in ordinary cases—namely six days as compared with forty-eight hours.

(146) Kolle (W.), Rothermundt (M.), & Peschié (S.). **Untersuchungen über die Wirkung von Quecksilberpräparaten auf Spirochätenkrankheiten.** [Investigations into the Actions of Mercury Compounds in Spirochaete Diseases.]—*Deut. Med. Wochenschr.* 1912. Aug. 22. Vol. 38. No. 34. pp. 1582-1585.

The authors' object has been to investigate the therapeutic and toxic effects of various compounds of mercury, and thus obtain data for the preparation of fresh compounds of high value for destroying spirochaetes but of low toxic value for the animal host.

A very large number of preparations have been tested. These are given in the form of a table and the following classes of compounds are included. Soluble and insoluble inorganic compounds of mercury; Soluble and insoluble organic compounds, compounds of the aliphatic and aromatic series being included in each group; Compounds of mercury with albumin and its cleavage products, both soluble and insoluble; Colloidal mercurial compounds.

(147) Levaditi (C.). **Intervention de l'Organisme dans la Guérison médicamenteuse des Maladies à Spirilles.** [The Part played by the Body in Recovery from Diseases caused by Spirilla brought about by the Administration of Drugs.]—*Bull. Soc. Path. Exot.* 1912. July. Vol. 5. No. 7. pp. 524-544. With 9 curves.

The author reviews briefly the work already published in connection with this subject and then gives details of a number of investigations upon which he bases the following conclusions:—

The rapidity with which the blood is sterilised varies inversely with the period elapsing between the administration of the drug ("606") and the time at which the crisis of the disease would naturally occur. A relatively rapid recovery may result from the administration of smaller doses of the drug if it be given during the pre-critical period of the disease. This conclusion does not apply, of course, to diseases which do not terminate in a crisis.

The author has found that in the case of rats infected with nagana, in which there is no crisis, the late administration of "606" does not produce more rapid recovery, but that if recovery takes place it is actually slower than when the drug is given in the early stages of the infection.

Two hypotheses may be given to explain the advantage of administering the drug during the pre-critical stage of the disease.

In the first place it may be supposed that, as the disease progresses and the crisis approaches, the vitality of the spirilla becomes

modified in such a way as to render them more sensitive to the microbicidal action of "606." It is known that the organisms present in the blood shortly before the crisis have a greater tendency to agglutinate, and survive *in vitro* for a shorter time than organisms taken in the early stages of infection. In this case the more rapid disappearance of the organisms from the blood would be due to a change in the spirilla and not to any special assistance on the part of the animal. The author does not think that this view can be accepted for the following reasons. If there is an actual modification in the vitality of the parasites one ought to be able to inject a fresh animal with spirilla obtained during the pre-critical period, subject the animal to treatment immediately after, and observe the prompt disappearance of the organism. The author's experiments have shown that this as a matter of fact does not happen, but that a change of animal host is associated with the immediate disappearance of the favourable influence of the pre-critical period.

The second hypothesis, which appears to the author to be the more probable, is that if the rat treated during the pre-critical period recover rapidly it is because the animal has acquired during this period the power of destroying the spirilla more easily. In administering the drug at this opportune moment the crisis is forced on and the host employs its ordinary defensive powers for the destruction of the parasites. It has been shown that in the treatment of spirillosis in the rat there is phagocytosis of the parasites, but also that the same phagocytosis is observed during the crisis which terminates experimental recurrent fever.

It will thus be observed that there is a particular time during the course of the disease at which it is most beneficial to administer the drug.

(148) Noguchi (H.). **The Pure Cultivation of** *Spirochaeta duttoni*, *Spirochaeta kochi*, *Spirochaeta obermeiri*, **and** *Spirochaeta novyi.*—*Jl. Experimental Med.* 1912. Aug. 1. Vol. 16 No. 2. pp. 199-210. With 2 plates.

The medium employed by the author for the successful cultivation of these blood parasites is prepared as follows. Pieces of fresh sterile tissue, usually rabbit kidney, are placed in sterile test-tubes. A few drops of citrated blood from the heart of infected rats or mice are added, and then about fifteen cubic centimetres of sterile ascitic or hydrocele fluid are poured into the tubes. To some of the tubes a small quantity of sterile paraffin oil was added. The tubes were then incubated at 37° C. It is very important that the ascitic fluid should contain no bile, and should be capable of forming a loose fibrin in the tubes. Fluids which have been heated to 60° C. for half an hour are unsuitable, as also are fluids that have been passed through a Berkefeld filter. The addition of broth or sugar diminishes the value of the medium.

The maximum growth is reached in about a week. The organisms do not multiply in an atmosphere of hydrogen or *in vacuo*, and no growth is obtained at room temperature.

Subcultures should be made by inoculating fresh tubes with a half to one cubic centimetre of culture from the fourth day up to the day of maximum growth. The organisms can be kept for a number of generations operating in this way, and the pathogenicity of the spirochaetes is not lost, although there appears to be a tendency to attenuation after a large number of subcultures have been made.

Growth is slower in the tubes containing paraffin oil.

(149) Nakano (H.). **Ueber die Reinzüchtung der** *Spirochaeta pallida.* [The Pure Cultivation of *Spirochaeta pallida.*]—*Deut. Med. Wochenschr.* 1912. July 11. Vol. 38. No. 28. pp. 1333-1335.

The author has been successful in obtaining cultures of the organism in impure culture in a number of different media, and it was with the object of purifying such contaminated cultures that the technique described in this paper was devised.

A sterile glass cylinder measuring about 7·5 by 2·5 centimetres is partly filled with sterile horse serum. A bacterial filter is then inserted into the cylinder. Both the cylinder and the filter are closed with rubber stoppers. The apparatus so fitted up is placed on four consecutive days for four hours in a water bath at 58°. On the fourth day it is placed in a water bath at 65° for 30 minutes, the contents being gelatinised in this way. The apparatus is tested by incubating it at 37°. The contaminated culture is introduced into the filter and the apparatus again placed in the incubator at the body temperature.

The author has been able to obtain pure cultures of a number of strains of the spirochaete in this way, including primary cultures. In some cases the colonies have appeared outside the filter on the third day. Immediately colonies are observed subcultures must be made from them because, although the spirochaetes pass through the filter more rapidly than bacteria, the bacteria do eventually escape. A few details are given regarding the appearance of the colonies, and the structure and characters of the cultivated organisms.

(150) Proca (G.), Danila (P.), & Stroe (A.). **Sur l'Isolement des Spirochètes.** [The Isolation of Spirochaetes.]—*Compt. Rend. Soc. Biol.* 1912. July 26. Vol. 73. No. 27. pp. 235-236.

The method which the authors have found most successful is that of Sowade slightly modified. Having obtained a mixed culture containing a large number of spirochaetes, sub-cultures are made in a number of tubes containing pyrogallol serum coagulated in the vertical position at 80° C., the medium being stabbed in the upper third only. The tubes are thus left for 10 to 20 days at 37° C. In the interval the serum beneath the zone of liquifaction is examined to see if it contains motile spirochaetes. If such be found the liquified serum is pipetted off and the tube is broken,

several fresh inoculations being made with particles of serum containing the spirochaetes in pure culture.

In this manner it is comparatively easy to obtain cultures which are pure in the lower half of the serum. Sometimes, however, symbiosis of spirochaetes is encountered. Sub-cultures with the pure spirochaete in pyrogallol serum, in simple serum, or in serum diluted with a solution of gentian always remain sterile. The spirochaetes become immobile after the first 24 hours.

Spirochaetes isolated in the lower layers of pyrogallol serum multiply abundantly when in making sub-cultures the typhoid bacillus is added. The bacillus mesentericus can also be used provided the medium employed be gentian serum. If the bacilli are added after 24 hours, by which time the spirochaetes have lost their motility, no effect is obtained. The simultaneous inoculation enables one to obtain abundant growth in every tube. After ten days spirochaetes are to be found throughout the serum but no actual colonies are formed.

Liquid media such as serum or serum broth are unsuitable. However, abundant cultures can be obtained in liquid serum if ordinary agar in the proportion of one in seven or eight of serum be added, the melted agar being well mixed with the serum at the time of inoculation. The cultures are odourless. The spirochaetes used by the authors were obtained from syphilitic lesions of the vulva.

(151) Scherechewsky (J.). **Reinzüchtung der Syphilisspirochaeten.** [The Pure Cultivation of the Spirochaete of Syphilis.]—*Deut. Med. Wochenschr.* 1912. July 11. Vol. 38. No. 28. pp. 1335-1336.

Tubes of sterile horse serum are plugged and capped and then placed in a water bath at 57° C. The temperature of the bath is the gradually raised until it reaches 70° C. at which temperature the serum becomes gelatinised. Immediately this condition is reached the tubes are placed in an incubator at body temperature to determine their sterility. Papules are washed with boracic acid solution and then cut out with curved scissors, care being taken to cut sufficiently deeply. The pieces of tissue are placed in the culture tubes and gently forced down into the depth of the medium, which returns and covers them. The tubes are then capped and placed in an incubator at 37° to 40° C.

The spirochaetes owing to their motility penetrate the medium, while contaminating organisms grow around and above the piece of tissue. On the fifth or sixth day a canal is made through the medium with a sterile capillary tube provided with a rubber teat. This canal reaches down to the piece of tissue, and into it about 3 cc. of 70 per cent. alcohol are ejected. The alcohol is allowed to remain for ten minutes and is then replaced by sterile distilled water, which in its turn is replaced by sterile paraffin oil. The tubes are again capped and incubated for a further five days. A scratch is made on the tube below the level of the now sterilised contamination and the tube broken off. In the medium contained in this part of the tube the spirochaetes are found in pure culture.

LEISHMANIASIS.

(152) Jemma (R.). **Sulla Leishmaniosi del Cane nei Dintorni di Palermo.** [Leishmaniasis of the Dog in the Neighbourhood of Palermo.]—*Pathologica.* 1912. Aug. 1. Vol. 4. No. 90. pp. 466-467.

Reference is first made to a previous publication by the author in which he stated that he had examined 227 dogs in Palermo for Leishmania with negative results. In the present investigation the parasite was found in two dogs. In the first instance the infected dog was found in a house near to one in which there was a child infected with the disease. The parents of the diseased child had had their own dog killed on account of its very poor condition, and it is probable that it also was infected. The diseased dog was examined, preparations being made from the liver and the bone marrow. In both of these parasites were found in large numbers.

In the second instance the owners of the dog could not be traced. Microscopic examination of the liver and bone marrow showed numerous parasites.

Seven dogs in all were examined in the neighbourhood in which there are numerous cases of human leishmaniasis, but in two only was the parasite found.

Basile was associated with the author in the investigations.

(153) Marshall (W. E.). **Further Experimental Investigation into Sudan Kala-Azar.**—*Jl. R. Army Med. Corps.* 1912. Sept. Vol. 19. No. 3. pp. 276-280. With 1 plate.

Experimental Kala Azar in the Dog.—Four dogs have been infected, and it has been shewn that the dog can be infected experimentally from the monkey, from another infected dog, and from human cases. Young dogs appear to be more susceptible, and in them the disease runs an acute course. In two experiments attempts were made to transmit the disease from dog to dog by means of the dog flea, but without success. Ticks also failed to transmit the disease in one experiment.

Experiments to determine by what means the disease may be conveyed from monkey to monkey failed, save in one case in which the result was doubtful. In this instance, if there was actual infection, the louse, the flea, and the mosquito may have played some part in the transmission. The author has encountered one monkey that appeared to have a natural immunity.

(154) Nicolle (C.) & Conor (M.). **Quelques Expériences Pratiquées avec le Virus de la Leishmaniose Naturelle du Chien. Reproduction de la Maladie chez le Singe.** [Experiments with the Virus of Canine Leishmaniosis. Reproduction of the Disease in the Monkey.]—*Bull. Soc. Path. Exot.* 1912. June. Vol. 5. No. 6. pp. 351-355.

The virus with which the experiments were made was obtained from Yakimoff, it having been found in a dog early in 1911. The lesions presented by the dog were the following: Emaciation,

purulent discharge from the eyes, loss of hair on some parts of the skin, enlargement of the lymphatic glands, thickening of the pericardium, congestion of the bone-marrow. Parasites were found in large numbers in smears from the bone marrow, few in the spleen, which was not enlarged, and none in smears from the liver, kidneys, lungs, or lymphatic glands.

Cultures were obtained, but they were contaminated, and subcultivation failed.

Two fresh dogs were inoculated successfully from the first animal. One of these died accidentally, but was found to be severely infected. No fresh dogs were inoculated from this animal.

The second dog was killed in December, 1911. The animal was fat and showed no gross lesions. On microscopic examination the parasite was found to be scantily present in the spleen, liver, and marrow. Cultures were obtained from the spleen on NNN medium, and subcultures were successful.

From this dog three further dogs and two monkeys were inoculated. In two of the dogs, which were old, the inoculation failed.

The third dog was killed about nine weeks after inoculation. There were no gross lesions, but parasites were found to be numerously present in the spleen, and bone-marrow, scanty in the liver and mesenteric glands, and none were found in the lungs, kidneys and blood. An attempt to inoculate another dog from this one failed.

One of the monkeys, an adult, failed to become infected. The second, which was very young, was suddenly seized with illness and showed symptoms of suffocation about six weeks after inoculation. It was killed on the evening of the same day when in a moribund condition.

Nothing was found to account for the suffocation. The heart was enormously dilated, but there were no lesions either of the myocardium or the valves. The spleen was enlarged.

In smears from the spleen parasites were fairly numerous; both intra- and extracellular forms being encountered. There were very few in the marrow and liver, and none were found in other organs.

Attempts to transmit the infection from this dog to another dog and a monkey failed.

The cultures obtained had up to the time of writing been carried on through twelve generations on NNN medium, and the characters presented were exactly those of cultures of the human parasite.

Two intraperitoneal inoculations with large quantities of culture into a monkey failed to set up infection.

(155) Yakimoff (W. L.) & Kohl-Yakimoff (N.). **L'Infection des Animaux de Laboratoire par *Leishmania infantum* Ch. Nicolle. (Deuxième Note Préliminaire.)** [The Infection of Laboratory Animals with *Leishmania infantum*.]—*Bull. Soc. Path. Exot.* 1912. June. Vol. 5. No. 6. pp. 355-357.

Infection of dogs by cultures.—The first dog was twice inoculated intravenously with cultures at an interval of three weeks. About four months later bone marrow was obtained by trephining

and Leishmania was discovered. A fortnight later the dog was killed and parasites were found to be fairly numerous in smears from the spleen and bone marrow, but none were found in smears from the liver, lungs, and lymphatic glands. A culture was obtained on NNN medium.

A second dog, inoculated intravenously with four tubes of culture a few days after the first, died on the forty-eighth day. Parasites were found to be present in the marrow in small numbers.

Infection of mice and rats with materials from infected organs.—The results of the inoculation of eleven mice and one rat are given. From these it is seen that in every mouse save one parasites were found in smear preparations from various organs. In one mouse and the rat microscopical examination was negative, but in both instances cultures were obtained from fragments of the spleen.

(156) Wenyon (C. M.). **Some Recent Advances in our Knowledge of Leishmaniasis.**—*Jl. London School of Trop. Med.* 1912. March. Vol. 1. Part 2. pp. 93-98.

Dogs have been found liable to Kala Azar in practically all the great centres of the infantile form of the disease—Tunis, Algeria, Lisbon, Malta, Sicily, Rome, and Greece. It has been suggested by Nicolle that the dog, in which the disease is mild and chronic, may act as a reservoir for the virus, which is transmitted to man by some biting arthropod. Basile claims to have proved that the transmission is effected by the dog flea (*Ctenocephalus canis*) and the domestic flea (*Pulex irritans*). His experiments were carried out on the following lines. First, healthy dogs introduced into houses where the disease existed became infected: fleas from infected houses in Sicily were taken to Rome and placed upon healthy, isolated dogs, which contracted the disease: dog fleas fed upon the spleen-juice of an infected dog were found to become infected with the cultural form of the organism. The gut-contents of fleas are infective to healthy dogs. Gabbi failed to confirm these results and is inclined to incriminate the mosquito on the grounds of positive results obtained by Franchini. Marshall, in the Sudan, has observed infection of a healthy monkey living in a cage with diseased ones. The organism is inoculable to the dog and monkey, and successful inoculations have been made into guinea-pigs, rats and rabbits. In the monkey the disease resembles that seen in children. A few experiments have been made in India to inoculate dogs but without success.

A certain amount of evidence has been adduced by Nicolle and Manceaux that dogs that have recovered are immune.

Cultures of the parasite of Oriental sore, first obtained by Nicolle on blood-agar, retain their virulence. Most observers are agreed that the house-fly may play some part in the transmission of the disease, and Row has found that the parasite may remain infective for monkeys for three hours after having been taken up by the fly. Wenyon has found that bed-bugs and

Stegomyia fasciata would feed on sores and take up parasites, and that in their guts the parasites develop into flagellate forms as they do in culture.

Donovan examined more than a thousand pariah dogs in India but failed to find infection, but possibly such dogs are immune.

RABIES.

(157) Bouffard (G.). **Sur l'Existence de la Rage Canine dans le Haut-Sénégal et le Niger.** [Canine Rabies in Senegal and the Niger.]—*Ann. Inst. Pasteur.* 1912. Sept. 25. Vol. 26. No. 9. pp. 727-731.

While there is no record of the occurrence of rabies in the human subject in the French colonies in West Africa, there would appear to be a disease of the dog the symptoms of which are strongly suggestive of rabies. It also appears to be certain that the bite of such dogs is fatal to other dogs, but without effect upon human beings.

The first animal suffering from this disease came into the author's possession in 1906. Unfortunately it escaped before any examination could be made, but not before it had bitten two other dogs belonging to the medical establishment.

One of these dogs sustained only a small bite on the paw which healed promptly, the dog remaining healthy for a year. The other dog was badly torn above the left eye. The wound was dressed and within a week healing was apparently complete, but on the 11th day the animal began to show peculiar symptoms. There was loss of appetite and an absence of desire to move, and throughout the following day the animal remained in a corner. About 6 o'clock in the evening it got up and began to rush about its enclosure in a state of great excitement. The author kept the animal under observation for an hour and noticed that from time to time it appeared to reel and to be uncertain in the movements of its hind legs. The next morning it was found dead in its cage.

Nothing interesting was found at the post-mortem save that the stomach was empty.

A fragment of the medulla was used for the inoculation of a rabbit, but the rabbit died on the fifth day from some accidental cause and its medulla was found to be non-infective for other rabbits. The strain was thus lost.

The following year while investigating sleeping sickness on the Bani River the author encountered a chief who informed him that there was a very fatal disease amongst dogs in his district. The affected dogs appeared to be mad, their bite was fatal to other dogs, but did not appear to cause any harm to human beings. This chief had no knowledge of such a disease occurring in man. Unfortunately the author was not in a position to inoculate animals and get the strain.

In August, 1909, the author came into possession of a dog at Kouloubah showing all the symptoms of dumb rabies. The animal was killed at once and its medulla used for the sub-dural inoculation of a rabbit.

For fifteen days this rabbit appeared to be in perfect health but on the sixteenth day it was found lying in its cage and when made to move it was observed that there was evidence of paralysis of the hind-quarters. The next day the paralysis was complete and the animal lay on its side showing marked dyspnoea. Death took place the same evening.

The medulla of this rabbit was used for the inoculation of two fresh rabbits.

During the next eight months thirteen passages were made, the period of incubation of the disease averaging about fifteen to twenty days. In two cases the period was twenty-five and in one case thirty-eight days.

The symptoms in every case were those already mentioned and in three cases there were additional symptoms of a furious type which made their appearance from twelve to twenty-four hours before death.

At each passage two or three animals were used and it was observed that about one animal in every six appeared to be refractory.

The author's successor continued the inoculations, but used one rabbit only at each passage with the result that the strain died out at the twenty-first passage, this animal remaining healthy.

This rabbit was used five months after the inoculation for a vaccine lymph control and two days later became paralysed and died shortly after.

Three further passages were made, the period of incubation being ten days in each of the first two passages but the third animal inoculated proved refractory and the strain was again lost.

(158) Harris (D. L.). **Recherches sur les Propriétés du Virus Rabique conservé a l'Etat Sec.** [The Properties of Rabic Virus preserved in the Dry Condition.]—*Ann. Inst. Pasteur.* 1912. Sept. 25. Vol. 26. No. 9. pp. 732-735.

The author's object was to devise some method of preserving the rabic virus which would obviate the difficulties attaching to the method devised by Pasteur, in order that a quantity of material might be kept at hand in places where vaccination is required only occasionally.

It has been shown by a number of authors that the virulence of the infective material is retained if a thin layer be made and then dried rapidly by means of sulphuric acid in vacuo. The amount of material that can be preserved in this way is too small to be of any value for vaccination purposes.

In conjunction with Shackell the author has described a method of preserving entire brains and cords in which the materials are frozen before being dried. Briefly the plan is as follows:—

The brain is first frozen with a mixture of ice and salt, and then without allowing it to thaw it is dried in vacuo with sulphuric acid.

Numerous experiments showed that about 1 per cent of the original virulence was retained in this way.

The authors believe that the desiccation of the cords according to PASTEUR'S procedure causes a progressive concentration of salts and other substances normally contained in the cord and that the destruction of the virus is proportional to this concentration.

During his experiments HARRIS has found that the degree of virulence retained in a cord preserved in this way depends upon the rapidity and completeness with which the material is frozen, and that it is possible to preserve from 30 per cent to 50 per cent. of the virulence. The method is as follows.

The brain or spinal cord is ground up in a porcelain mortar and water is added drop by drop until a thick homogenous paste is obtained. A little carbonic acid snow is then added slowly and with constant stirring in order to avoid freezing the material into a solid mass. When freezing is complete the material is as fragile as glass and can be easily pulverised. It is necessary to add a little of the snow from time to time to prevent thawing.

The mixture is then immediately transferred to a cold vessel and placed in the bottom of an exsiccator which has been previously embedded to half its depth in a mixture of ice and salt at a temperature of —18° C. There is placed in the upper part of the exsiccator a vessel containing sulphuric acid in such a manner that there is free circulation of air between the vessel containing the cord and the exsiccator. Solidification occurs if the acid be placed too near the mixture. There should be a vacuum of 2 mm. of mercury, and the exsiccator should be gently agitated from time to time in order to mix thoroughly the absorbed water with the acid.

A brain can be desiccated in this manner in from thirty-six to forty-eight hours, but towards the end of the operation the temperature must not pass —10° C.

The dried product forms a very light hygroscopic powder which absorbs water from the air up to 3 per cent and then becomes soft, and within a few hours loses all its virulence. To protect the material from moisture it is sealed up in tubes.

It has been shown in numerous experiments that 0·00002 g. of cord dried in this way produces rabies in rabbits on the 6th day. Half this quantity kept at 8-10° C. for a month and protected from light is also capable of setting up the disease. After two months the virulence is diminished by half.

Phosphoric acid is less satisfactory than sulphuric acid for the desiccation.

Rabbits and dogs can be rapidly immunised with this material by HÖGYES' method, and the author has immunised dogs with cords preserved for three months.

Further experiments are in train to ascertain the effects of certain chemical substances, temperature, light, etc. on the loss of virulence.

The advantages claimed for the method are, that the powder can be easily weighed, its properties can be determined exactly, and that it is relatively permanent.

HELMINTHS.

(159) Connal (A.). **Some Nematode Worms from Lagos.**—*Jl. London School Trop. Med.* 1912. July. Vol. 1. Part 3. pp. 229-237.

The six species of worms described were found in a collection made from various small wild mammals, birds, and reptiles, by Beale-Brown in Lagos.

Strongyluris streptoesophageus n. sp. Found in large numbers in the stomach and intestine of the common West African lizard, *Agama colonorum*.

Male. Length 8·6 mm. Breadth 0·44 mm. Body tapers gradually to the head end. Tail perfectly straight. Cuticle smooth. Mouth provided with three simple lips. The oesophagus shows a sharp kink near the anterior end, and behind this the tubes show three bends. There is a well-marked bulb. There are slender subcuticular papillae along the whole length of the body. The anus is quite close to the tail. The spicules are long, equal in length, and tapering. There is a sucker ventrally placed 0·76 mm. from the tail. There are three pairs of pre-anal and six pairs of post-anal papillae.

Female. Length 9·1 mm. Greatest breadth 0·5 mm. Vulva 3·33 mm. from the tip of the tail. Oviparous, the ova measuring 0·06 by 0·03 mm. The sucking disc is absent. Anus 0·2 mm. from the tip of the tail. The species appears to be closely allied to the *Strongyluris brevicaudata*.

Filaria yaba. n. sp. Found in large numbers in the thoracic cavity of *Centropus senegalensis*.

Male. Length 19 mm. Greatest breadth 0·6 mm. Extremities taper somewhat abruptly. Tail bluntly pointed and coiled at least 1½ times. Skin smooth. Mouth simple with a minute papilla on the outer aspect of each of the three lips. Oesophagus somewhat bottle-shaped. Anus about a tenth of a millimetre from the tail. Spicules short and boomerang-like in shape. One pair of pre-anal and four pairs of post-anal papillae, all small.

Female. Length 23-33 mm. Greatest breadth 0·8 mm. Both ends bluntly pointed. Vulva about 1 mm. from the head. Ovoviviparous. Embryos much coiled. Anus nearly terminal.

Triplotriaena falconis, n. sp. Found in large numbers in the thoracic cavity of a hawk (sp. ?).

Male. 30·1 mm. Greatest 0·46 mm. Tapers at both ends. Cuticle finely striated. Mouth has three simple lips devoid of papillae. Oesophagus slender. Spicules of unequal length. Larger one straight, blunt pointed, and tapering. Shorter one corkscrew-like with four distinct twists, more pointed than the long one and sligtly narrower. No anal papillae. Anus nearly terminal.

Female. Length 55 mm. Greatest breadth 0·6 mm. Vulva very close to the anterior extremity. Ovoviviparous. Anus nearly terminal.

Ascaris rosarius n. sp. Found in the stomach of a small grey heron.

Male. Length 26 mm. Greatest breadth 0·6 mm. Anterior end of the worm cylindrical and narrow for a distance of 0·7 mm.

then broadens abruptly. Posteriorly the body tapers rapidly to a sharp point. Cuticle strongly striated, and numerous regularly arranged refractile dots give a beaded appearance to it. Tail coiled ventrally. Mouth provided with six fleshy lips, three large and three small placed alternately. Distinct collar of cuticle around the base of the lips. Oesophagus gradually widens, and there is a blind prolongation of it about 0·5 mm. in length running alongside the intestine. There is a blind prolongation of the intestine forwards alongside the oesophagus. Anus 0·22 mm. from the tip of the tail. Three pairs of post-anal papillae. Spicules long, equal in size and shape, and have a flanged appearance. Numerous pre-anal papillae.

Female. Length 30 mm. Greatest breadth 0·7 mm. Vulva 1·5 mm. from head. Oviparous. Shell of the eggs thick.

Spiroptera deflecta. n. sp. Found in the stomach of the palm squirrel.

Male. 19·7 mm. Greatest breadth 0·6 mm. Body tapers to a fairly sharp point anteriorly. The anterior portion bent laterally at a distinct angle. Tail tapers more quickly than the anterior end, and shows a marked curve. Cuticle finely striated. Mouth has six small lobular lips. Oesophagus distinctly bulbous at its junction with the intestine. The opening of the oesophagus into the intestine is furnished with three valvular flaps depending from a collar-like thickening. Anus situated 0·4 mm. from the tail. Spicules short and broad, and unequal in size and shape. The dorsal part and the tip of each is thickened, while the thinned and broadened ventral portion presents a frescoed appearance. There is a curved accessory piece. At the tail end on the ventral aspect there is a well-marked ala nearly 2 mm. in length. Five pairs of pre-anal and three pairs of post-anal papillae.

Female. Length 26 mm. Vulva situated about the middle of the body. Anus 0·3 mm. from tail.

Echinorhynchus centropi, Porta, 1910. Found in the intestine of *Centropus senegalensis.*

Male. Length 29 mm. Greatest breadth 1·2 mm. Body cylindrical but somewhat swollen towards the anterior part. Tail rounded. Cuticle smooth. Rostrum nearly globular. Hooks arranged alternately nine and ten in a row longitudinally, and twenty-six in a transverse row.

The five posterior hooks in a row of nine are smaller than the anterior four. The cirrus-pouch measures 3·6 mm. in length.

Female. Length 48 mm. Greatest breadth 1·5 mm. General characters like those of the male, but the posterior end of the body has a finger-like prolongation 0·34 mm. in length. Oviparous. Eggs thick-shelled and rather elongated.

(160) Innes (J. A.). *Gastrothylax bubalis,* **n.sp. With a Few Notes on the Genus** *Gastrothylax* **(Poirier).**—*Parasitology.* Sept. 1912. Vol. 5. No. 3. pp. 217-226. With 8 text-figures.

The parasite was found by Brown, British Central Africa, in the stomach of a Rhodesian hartebeest, and about one hundred and twenty specimens were secured.

The parasites were examined in serial sections, and it was found that sections, 10 microns thick, could be obtained if the process of embedding in paraffin were carried out rapidly. If the process occupied more than two hours the specimens were too brittle to cut well. The best results were obtained by staining the parasites in bulk with MAYER'S paracarmine for three days, and then differentiating in several changes of 70 per cent. alcohol to which a little 1 per cent. solution of ammonium chloride had been added. Imperfect fixation made it impossible to study the finer histological details. In tracing the ducts rough reconstructions were made.

The generic characteristic of the genus is the large atrium opening anteriorly by the atrial pore.

External anatomy. The parasite is conical in shape with a blunt anterior end. It measures from eight to ten millimetres in length and from three to four in diameter at its thickest (posterior) part. The parasite may be either expanded or contracted, and in the latter case the surface is covered with wrinkles. The posterior sucker in the expended specimens occupies practically the whole of the posterior part of the parasite, but in contracted forms its diameter is about one-third of that and it is retracted somewhat. The mouth opens anteriorly exactly at the apex of the cone and is surrounded by a mass of muscular tissue. Oral papillae are present.

The transverse opening of the genital atrium opens a little behind the mouth on the mid-ventral line, and it is lined with papillae.

The ventral side of the parasite is slightly concave or straight, the dorsal side being convex.

Internal anatomy.—The alimentary canal consists of a terminal mouth, a muscular pharynx, a short oesophagus which terminates in two blind pouches passing down on either side of the atrium, dorsal to the testes. The gut branches show varied convolutions. The atrium is large and expands posteriorly where it terminates just above the anterior boundary of the testes. It is triangular in cross-section, the apex of the triangle being ventral. The physiology of the organ is not known but it is possibly used as a receptacle for ova. It opens to the exterior on the ventral surface just behind the mouth, and in this region it also receives the genital opening. The reproductive system is almost entirely included between the base of the atrim and the posterior sucker.

The male system consists of a pair of large round testes, which do not extend close to the body-wall of the parasite, nor into the atrial cavity, two vasa efferentia, and a single long vas deferens opening along with the vagina at the genital aperture. The vas deferens and the vagina run dorsal to the atrium.

The female system consists of ovary, shell gland, oviduct, uterus and vagina. The shell gland and the ovary are very close together, and the duct from the shell gland almost immediately joins that of the ovary. These organs are situated between the testes. The ova measure about one hundred and ten by sixty microns.

The excretory vesicle which is very irregular in outline lies between the shell gland and the posterior sucker, its canal is short and does not unite with Laurer's canal.

The muscular tissue of the parasite appears to be concentrated about the suctorial regions.

The parasite is believed to be non-pathogenic.

Notes on the genus Gastrothylax.

An outline of the historical details connected with the genus is given, and also a description of the general features characterising the genus. Finally the distinguishing characters of the ten species comprising the genus are tabulated.

(161) Jowett (W.). **Nodular Intestinal Disease of Cattle.**—*Journ. Comp. Path. & Ther.* 1912. March. Vol. 25. No. 1. pp. 15-22. With 5 figures.

The investigations are based upon the examination of material that the author has obtained from cattle during post-mortem examinations. He has frequently encountered the disease in cattle in South Africa. The disease is characterised by the formation of nodules in the sub-mucous coat of the intestine, the nodules varying in size from that of a pin's head to that of a pea. The lesions may be scanty or very numerous. It is only rarely that there is any ulceration of the mucous membrane over the nodules, but there may be considerable thickening. The lesions are more frequently encountered in the small than in the large intestine. The colour of the nodules varies with age. The young lesions are as a rule haemorrhagic, the medium-sized are either black or white and black, and the larger or older nodules are either uniformly white or yellowish, and they may have greenish yellow contents. The oldest lesions are for the most part fibrous, and contain caseous or calcareous material.

It is difficult to demonstrate the parasite which is responsible for the condition in any save young lesions. The causal parasite is the embryo of a nematode worm which measures from two to three millimetres in length.

In the nodules the worm is in the embryo stage, but after ecdysis, by which the worm is converted into an immature adult, it escapes into the lumen of the gut. The adult worms measure about two centimetres in length. In the intestine the worms become sexually mature and copulate. The eggs show segmentation while still in the uterus of the female, as is also observed in the closely related parasite of man.

The eggs are passed out and under suitable conditions undergo processes of development leading to the formation of embryos which gain access to a bovine host with either food or water.

It has been shown by Marotel that the causal parasite in an oesophagostome, and from the more recent researches of Guillé, Marotel, and Panisset it would appear that the worm responsible for disease in cattle is a distinct species and the name *Oesophagostomum biramosum* has been given to it. The

characters by which this worm is distinguished from the *O. radiatum* are: (1) the possession of a cervical swelling, somewhat hour-glass-shaped but non-vesicular; and (2) two distinct branches to each of the posterior ribs in the caudal bursa of the male, the outer division of each main branch of the dorsal ray being better marked and somewhat longer than is the case in *O. radiatum*.

PLAGUE.

(162) Dujardin-Beaumetz (E.) & Mosny (E.). **Évolution de la Peste chez la Marmotte pendant l'Hibernation.** [Plague in the Marmotte during Hibernation.]—*Compt. Rend. Acad. Sci.* 1912. July 22. Vol. 155. No. 4. pp. 329-332.

It has long been known that in Transbaikal and Mongolia there are persistent plague centres, and that trappers contract the disease from marmottes and tarbagans in these areas. It appears that the marmotte, like the rat and other rodents, acts as a reservoir for the plague virus.

Until comparatively recently it was believed that in countries where the rat is the transmitting agent, the disease persisted in that animal in a chronic form. It is now known, however, that such is not the case, and that for the survival of the virus there must be a sufficient number of rats to assure the continuous transmission of the disease from rat to rat.

In Mongolia where the winter is severe there could be no question of the virus persisting outside the animal body because it is very susceptible to external influences. It therefore had to be supposed that the virus was maintained either in the flea or in the marmotte, since rats are not found in these plague areas.

The experiments of Gauthier and Raybaud have shown that the bacillus can retain its vitality and virulence in the stomachs of fleas for 45 days in an ice chest, but it was not shown whether such fleas were capable of transmitting the disease by biting after such a period.

It has already been shown that hibernating marmottes resist infection with tuberculosis, and that trypanosomes fail to infect provided a period of five days elapses before hibernation terminates. In some experiments carried out by Wurtz marmottes were inoculated with plague, but owing to the conditions being unfavourable for hibernation the animals awoke and death occurred some days after inoculation.

The authors have made three experiments with marmottes captured during hibernation. The three animals were kept together at a constant temperature of 5-10° C., and were protected from all stimuli likely to wake them.

The first animal was inoculated subcutaneously with virulent plague virus. It was subjected to handling on a number of occasions causing it to wake, but the animal survived till the 61st day having lost about 12 per cent. in weight during the interval.

The second animal was inoculated in a similar manner, but was not handled more than was absolutely necessary. This animal survived 115 days and died after waking at the normal time.

The third animal was kept as a control to show the susceptibility of marmottes to the virus of plague in the waking state. This animal died two and a half days after inoculation.

The possible explanations are that either the virus persists without multiplication, or that multiplication is very much restricted during hibernation, and becomes very rapid immediately the period of hibernation terminates.

(163) Matsuo (K.). **Gleichzeitiges plötzliches Auftreten von Pestfällen bei Menschen und Eseln in demselben Gehöft.** [The Simultaneous and Sudden Occurrence of Cases of Plague in Man and Donkeys.]—*Centralb. f. Bakt.* 1. Abt., Orig. 1912. Aug. 10. Vol. 65. Nos. 6-7. pp. 417-423. With 1 plate.

The outbreak of pneumonic plague which occurred in Northern Manchuria in October, 1910, and which lasted till April, 1911, was of extraordinary severity and claimed more than 40,000 victims. The author was stationed at Mukden in January, 1911, for duty in connection with the outbreak. Towards the end of February information was received that a donkey had died of plague in Fushun, and shortly after a dog was found to have died from the disease in Changchun.

When the outbreak was beginning to die out the author accidentally encountered a case of the disease in a donkey, and by following up the case discovered a further instance of infection in the donkey in which nearly every man and donkey in a particular building became infected one after the other. The circumstances of the case were as follows:—

A miller in Mukden employed eleven men and kept twelve donkeys. In March one of the men died, plague being particularly prevalent in Mukden at the time. The employer attempted to conceal the death, and placed the body in a loft where it was subsequently found, and plague was proved to have been the cause of death.

Soon after the man's death one of the donkeys became ill, losing its appetite and coughing severely. The animal was sold shortly after and its purchaser could not be traced. After a short interval another man died. Subsequently seven donkeys became infected one from another. Of these three were sold and four died. Two healthy donkeys were sold at the same time as the diseased ones, but none of the animals nor the purchaser could be traced. One of the dead donkeys was removed and buried. Before the other three dead donkeys could be removed the miller and all his employees were placed in the isolation hospital, the donkeys being afterwards buried.

The two remaining animals were isolated and remained healthy. The miller and seven of the nine men died of plague.

The author received instructions to examine the three donkeys buried. The following lesions were found in the first of the three: The brain was putrid but showed congestion, as did also the medulla, there were no adhesions of the pleura, but the lungs were congested, and there was distinct infiltration of the upper border of both lungs. The heart was hypertrophied, liver three times the

normal size, spleen reduced in size with numerous wrinkles, stomach normal, hyperaemia of a portion of the large intestine with petechiae, kidneys normal.

Only the lungs were removed from the other two carcases, and there was marked infiltration of the borders of the lungs and distinct hepatisation of the right lung.

Fluid was obtained by puncture of the lungs and from the upper air passages and bacilli resembling the plague bacillus were found in addition to numbers of other bacteria—principally streptococci. Smear preparations were made from the various organs and stained by Loeffler's method. Bacilli were found to be numerously present in smears taken from the lungs, scanty in smears from the liver, and absent in smears from the heart-blood, spleen, brain, and kidneys.

Nine mice were inoculated with materials taken from various organs, and practically all died in 40-45 hours, a single one surviving till the 72nd hour. The organisms isolated from the mice agreed exactly in cultural and staining characters with the bacillus of plague.

Owing to the impossibility of getting experimental animals only three experimental inoculations could be made to test the virulence of the bacilli cultivated from the mice. Control inoculations were made with sputum from a case of plague and the results were in absolute agreement, a guinea-pig and a mouse inoculated in each instance with 1/100,000 of a loopful dying in 75-90 hours. Plague bacilli were demonstrated in the blood, liver and spleen of the experimental animals. Two further guinea-pigs inoculated with 1/1,000,000 of a loopful were still healthy some months after.

Immune plague serum (horse) was used to compare the agglutination of the organisms isolated from the donkeys with controls of plague bacilli. Here again the results were in absolute agreement, the serum causing agglutination with both strains of organisms in dilutions up to 100 but not higher.

MISCELLANEOUS.

(164) Lheritier (A.), Fleury (A.), & Tribout (A.). **Moutons Algériens et Bactéridie Charbonneuse.** [Algerian Sheep and the Anthrax Bacillus.]—*Bull. Soc. Path. Exot.* 1912. June. Vol. 5. No. 6. pp. 336-339.

After a brief reference to an outbreak of anthrax among some cattle and to one case in a sheep, the authors give a short review of the experiments carried out by Chauveau which indicated that Algerian sheep are more or less resistant to the bacillus of anthrax as it occurs in Europe.

Experiments were made with the object of comparing the virulence of the bacillus obtained from an Algerian sheep dead of the disease with that of a strain of the organism obtained from Europe.

In a preliminary experiment it was found that whereas the Algerian strain proved fatal to a guinea-pig in 33 hours, the European strain did not cause death till the 40th hour, the guinea-pigs used being of the same weight, and a similar dose being administered in each case.

In the second experiment Algerian sheep were used. These were divided into two lots of five. One batch was inoculated with the Algerian strain and the other with a French strain. In each series four were inoculated subcutaneously and one intravenously, the doses being strictly comparable.

The whole of the sheep inoculated with the Algerian strain died, while the whole of those done with the French strain recovered.

The sheep which had resisted inoculation with the French virus were inoculated three months later with a large dose of the Algerian strain to ascertain whether they had acquired any immunity. One fresh sheep was inoculated at the same time as a control. The control sheep died, but none of the others showed any symptom save a slight elevation of temperature.

(165) Leboeuf (A.). **Existence de Lepra murium (Lèpre des Rats) en Nouvelle-Caledonie.** [Leprosy of the Rat in New Caledonia.]—*Bull. Soc. Path. Exot.* 1912. July. Vol. 5. No. 7. pp. 463-465.

Having given a brief review of the literature the author states that he has found three infected rats out of ninety-nine examined. He agrees with other investigators that the bacillus found is easily distinguishable from the bacillus of Hansen. The organism is longer, as a rule stains uniformly, is slightly curved, frequently shows a spherical swelling at one end, and shows no tendency to collect into rounded masses.

The three rats were all adults and appeared to be in perfect health.

In the first acid-fast bacilli were encountered in the axillary and inguinal glands on the left side, and a few in the apex of the left lung.

The second rat had very large axillary and inguinal glands on both sides of the body and bacilli were very numerously present.

The third rat showed fairly numerous bacilli in the left inguinal gland, and they were not scanty in the left axillary gland and in both glands on the right side.

The author points out that in one animal only was there any evidence of invasion of the viscera, and that in the same animal the glands were less seriously affected than in the others.

(166) **Distribution, Etiologie et Prophylaxie de la Fièvre Ondulante.** [Malta Fever.]—*Bull. Off. Intern. d'Hyg. Pub.* 1912. July. Vol. 4. No. 7. pp. 1180-1211.

This paper gives a brief review of the work done in connection with this disease up to date and deals with the following points: Preliminary considerations, including synonyms; Historical; Distribution; Symptomatology; Prognosis and Evolution; Epidemiology; Etiology and the Biological Characters of the causal organism; and, finally, Prophylaxis. From the veterinary point of view the principal interest centres in the details that are given regarding the extent to which goats are affected in various places,

and the occurrence of infection in animals of other species than the goat. In connection with the occurrence of infection in goats the following table is given:

Authors.	Place.	Percentage of goats infected.
Zammit and Horrocks	Malta	50
Sergent	Algeria	3·4
Nicolle and Conseil	Tunis	30·7
Conor and Huon	Marseilles	34·2
Aubert, Cantaloube & Thibault	Gard	29·0
Shaw	Malta	9·9

The goat is far more frequently affected than other species of animals, but epidemics are not all unknown in such animals.

The sheep appears to be next most commonly infected after the goat, and as in the goat abortion is very commonly observed.

The organism has been isolated from the mule, donkey and horse in Algeria. A number of authors have recorded its occurrence in cattle, and a number of birds are susceptible. The susceptibility of the rabbit is not yet fully established.

(167) Bevan (Ll. E. W.). **Ephemeral Fever, or Three Days Sickness of Cattle.**—*Veterinary Jl.* 1912. Aug. Vol. 68. No. 446. pp. 458-461.

This paper contains a brief summary of what is known regarding this disease, emphasising some points by reference to particular cases. The disease is remarkable in that outbreaks occur in areas far removed from each other, and between which there has been no interchange of stock. The disease attacks animals of all ages, conditions, and sexes, and animals immune to red-water, anaplasmosis, and Coast fever possess no immunity. The onset of the disease in a herd is sudden, and within a day or two a number of animals may be affected. In Cape Colony the disease has been observed in stable animals. It is known that the virus of the disease is present in the blood of a sick animal, and proof has been furnished that it can be transmitted to other animals by blood inoculation. The period of incubation in such cases is two or three days and the attack is followed by immunity which lasts for about six weeks. The blood of a recovered animal does not transmit the disease. It has been suggested that the disease is insect transmitted.

Typical cases run their course in about three days, but in complicated cases the period of illness may last for weeks. Acute cases have been known to recover within twenty-four hours. The condition must not be confused with "Lamziekte" which is caused by eating *Crotalaria burkeana*.

The stiffness may be observed in any or all the legs, and it may pass rapidly from one limb to another. The muscles of the neck or the back may be involved. Very often ropy saliva hangs from the lips and discharge pours from the nose. The tissues around the eyes may be very swollen. There is marked elevation of temperature, and constipation is frequently observed.

The mortality is low and it is difficult to say what are the lesions proper to the disease.

(168) Schellhase (W.). **Ein Beitrag zur Kenntnis der ansteckenden Lungenbrustfellentzündung der Ziegen in Deutsch-Ostafrika.** [Contagious Pleuro-pneumonia of the Goat in German East Africa.]—*Zeitschr. f. Infektionskrankh., Parasit. Krankh., u. Hyg. d. Haust.* 1912. Vol. 12. No. 1. pp. 70-83.

The author, who carried out his investigations in the field without laboratory equipment, arrives at the following conclusions:—

1. The disease is transmissible to healthy goats by intrapulmonary inoculation with lymph obtained from the lung tissue of a diseased animal.

2. The disease is not transmissible by subcutaneous inoculation with the same material.

3. The disease cannot be transmitted to calves by intrapulmonary inoculation.

4. Intrapulmonary inoculation fails to transmit the disease to sheep, but the inoculation causes a localised inflammation of the lung tissue.

5. Subcutaneous inoculation of goats with lung lymph causes a local and a general reaction. The disease cannot be transmitted by intrapulmonary inoculation to sheep so treated.

6. Smears from the lung tissue of natural and experimental cases show large numbers of coccus-like bodies which are easy to stain, but the significance of which could not be determined.

(169) Bridé (J.), Nègre (L.), & Trouette (G.). **Recherches sur la Lymphangite Épizootique en Algérie.** [Epizootic Lymphangitis in Algeria.]—*Ann. Inst. Pasteur.* 1912. Sept. 25. Vol. 26. No. 9. pp. 701-726. With one plate.

The authors deal at some length with the clinical aspects of the disease before passing to the consideration of the pathological anatomy of the condition and the causal organism.

Examination of an excised corded lymphatic vessel shows that in the smallest nodules the contents are of a greyish red colour, whereas in the larger and older lesions they approximate more and more to pus in appearance.

Microscopic examination of one of the smallest nodules shows that the contents appear to consist of a colony of cryptococci, many of which are multiplying; but very few leucocytes are to be seen.

On examining larger lesions it is found that leucocytes have gained access to the mass of parasites and that the number of organisms showing evidence of multiplication is smaller. It is further observed that a number of the organisms have been ingested by phagocytes.

The organism can be stained by Gram's method, but a large proportion of the elements are decolourised. Better results are to be obtained by Claudius' method, but both the gentian violet and the picric acid should be allowed to act for an hour and chloroform should be used as the decolourising agent. Good results may also be obtained with the toluidine blue method of Dominici and with Giemsa's solution.

In sections it can be seen that the organisms invade the whole of the lymphatic vessel and a certain number are to be found free among the proliferating connective tissue cells.

The organism is not a parasite of the white corpuscles and the occurrence of elements within leucocytes is simply evidence of phagocytosis.

In the opinion of the authors the view that the cryptococcus is a protozoal parasite is a mistaken one, this view having been based upon the examination of lesions in too advanced a stage and in which phagocytosis was active. They state that when a lesion has become appreciable to the touch in a living animal it is already invaded by leucocytes and phagocytosis has commenced.

The disease must be considered as a local affection which spreads in the centripetal direction along the course of the lymphatics. The organisms rarely or never pass the glands at the entrance to the chest or the abdomen to determine lesions in the internal organs.

A brief review of the opinions expressed by various authors as to the nature of the organism is given. The authors disagree with the view that the double contoured envelope in an artefact, and that lemon-shaped forms are rare. They further disagree with the explanation offered by the partisans of the protozoal theory that budding forms represent two individuals joined together.

In support of their view that the parasite multiplies by a process of budding they state that in the young lesions described numerous parasites are to be found with daughter cells attached to them and showing a gap in the capsule through which the daughter cell is herniated. They further state that parasites may be observed having daughter cells of the second generation also attached to them.

The authors express no opinion as to the nature of the cell contents.

A large number of attempts have been made to cultivate the organism on the most diverse media both of animal and vegetable origin, but without success. In a few cases the authors have thought that there was a larger number of budding forms in certain media than in the seed material, but they have never obtained obvious growths, nor have subcultures ever proved successful.

This failure to cultivate the organism is not evidence of its protozoal nature, for Leishmania, to which some authors believe that the organism approximates, can be easily cultivated.

The authors believe that the causal organism is of the nature of yeast and with this idea carried out "deviation of the complement" experiments.

In the first instance the presence of sensitiser was sought in the serum of affected animals using a dilution of cryptococci in salt solution as antigen. The results indicated that the serum of infected animals contains a sensitiser for the cryptococcus.

In the second series of tests a culture of a known blastomycete was used in the place of the dilution of cryptococci as antigen.

The experiments were repeated with several kinds of yeast and with the sera of diseased and normal horses. The results obtained reproduced those obtained in the first series of experiments, positive results being obtained with a rice yeast.

In a third experiment it was found that a bacterium such as the *B. coli* in place of the yeast gave negative results.

A test was then made as to whether the cryptococcus or the yeast was capable of fixing any other sensitiser than that contained in the serum of an animal infected with epizootic lymphangitis. The result was again negative.

Control tests were made using an anti-yeast serum with the various kinds of yeast used in previous experiments. The anti-yeast serum was obtained from a rabbit which had received a number of injections either subcutaneously or intraperitoneally, of beer yeast. The tests were repeated with the serum of a normal rabbit. In every case there was no haemolysis in the tubes containing the anti-yeast serum and the different antigens, while in the ordinary control tubes and in the tubes containing normal rabbit serum there was haemolysis.

As a final test the serum of an infected animal was tested with cultures of *Leishmania infantum* and *Trypanosoma vespertilionis*, the antigens being cultures of these organisms obtained in Novy-McNeal-Nicolle medium. A similar series of tests was carried out with normal serum. The results indicated that these antigens fail to prove the presence of any sensitiser in serum from infected animals.

In the author's opinion these results appear to support, if not prove, the theory that the causal organism of epizootic lymphangitis is a blastomycete.

The authors believe that infection always takes place through a wound, but they find that it is impossible to transmit the disease with certainty by inoculation of pus. They do not agree with the view that the disease is insect transmitted.

One case is on record of transmission of the disease to the human subject, the infection taking place through a wound.

Treatment.—The authors give details of forty-three cases of the disease treated with "606."

After a few experiments they found that as good results were obtained with the comparatively small dose of 1 gramme as with larger doses. The drug was administered intravenously according to EHRLICH's directions. They have never observed any general disturbance save in one case in which a mule received 3 grammes and showed symptoms of slight colic and diarrhoea.

The effect of the drug is rapid. When the disease is recent and the primary lesion is situated on the middle portion of a limb (about the knee) there is rapid healing, the corded vessels diminish in size and buds that are already appreciable burst and become indurated.

If the disease has been in existence for some time and the initial wound is situated on the lower part of the limb the effect of the injection is in many cases to cause the appearance of fresh buds along the course of the diseased lymphatic vessel. To the inexperienced this might appear to be an aggravation of the

disease, whereas it is in reality a reaction on the part of the animal for the removal of the parasites. Three weeks or a month should be allowed to elapse before the results of the injection are judged.

In the table given, comprising forty-three cases, twenty-seven animals are said to have recovered, while seven are said to be on the road to recovery; the remainder having died or been killed. It is pointed out that in every case in which the animal had to be killed the primary infection occurred in the lower parts of the limbs.

When the diseased lymphatic vessel is well marked and is in a position which renders surgical interference easy this method of treatment should be resorted to, "606" being reserved for those cases that are inoperable.

The case of human infection terminated in complete recovery in three days after the intravenous injection of 0·6 g. of "606."

Prophylaxis.—Before the experiments with "606" were carried out hypodermic inoculations of yeasts were tried, to see whether they might not lead to the production of antibodies capable of exercising an effect on the cryptococci. It was found that there was increasing intolerance of the organism after each inoculation, abscess-formation with discharge of pus occurring within progressively shorter periods. The authors have attempted to make use of this fact in protecting animals in an infected stable by giving half of the animals an inoculation with yeast, leaving the others as controls. Sufficient time has not yet elapsed to enable any statement regarding this experiment to be made.

(170) Walker (G. K.). **The Treatment of Rinderpest and Haemorrhagic Septicaemia with Permanganate of Potash.**—*Jl. Comp. Path. & Therap.* 1912. Vol. 25. No. 3. pp. 185-202.

Rinderpest.

Originally the drug was tried upon two cases of the disease both of which recovered, the dose in each case being ½ dram dissolved in about a gallon of water. Shortly afterwards two further animals were treated, one of these receiving 1½ drams and the other ½ dram. Both of the animals recovered.

Some months later there was an outbreak of the disease in a herd of animals, eighty-eight of which were attacked. Of these 55 had died. Fifteen of the survivors were found to be suffering from the disease, but owing to opposition on the part of the owners it was found impossible to subject all to the treatment. Six were treated, however, the remainder acting as controls.

Subsequently twenty-four additional animals were brought under treatment. Of the thirty animals treated only three died, while of thirteen controls seven died.

The dose of the drug given varied according to the size and age of the animal and the clinical condition. In one or two instances a dose of ½ dram was administered, but as a rule the dose was either 1 or 2 drams. Some of the animals received a single dose only,

while others received as many as four, and in one instance six. The drug was dissolved in about half a gallon of water with an ounce or two of vinegar added.

Post-mortem examinations were made in all the fatal cases and the diagnosis confirmed.

Haemorrhagic Septicaemia.

Three animals were originally treated, with two recoveries, the third animal being in a critical condition at the time of treatment.

Twenty-three cases in all were treated and recovery followed in ten of these. Since the mortality usually lies between 90 and 100 per cent., the results appeared encouraging.

The percentage of recoveries in cattle was very much higher than in buffaloes (83·3 per cent. as compared with 29·4 per cent.). The greater susceptibility of the buffalo indicates that the larger doses should be tried.

The drug was administered as in the cases of rinderpest. The following is an abstract from the conclusions drawn by the author:—

While no definite conclusions can be drawn from the experiments described they appear to justify the hope that the treatment may prove useful. It is probable that considerably larger doses can be given and with good effect. Further experiments are required to ascertain the maximum dose that can be tolerated.

Calves may be given ½ to 1 dram, medium-sized cattle 2 drams, and animals weighing over 500 lbs. 3 to 4 drams.

It has yet to be decided whether the best results are to be looked for from the administration of a large initial dose or from the daily administration of moderate doses. The former would be the more convenient and in the case of haemorrhagic septicaemia there is rarely time for more than one dose.

The method will have to be tried in cases of rinderpest among animals having little or no natural immunity.

Hypodermic injection of the drug might be tried, but it is probable that its caustic nature would be injurious to the tissues. The injection of the drug direct into the abomasum is worthy of trial.

(171) Nicolas (C.). **Observation Clinique d'une Affection Chevaline sevissant à Nindiah et Nindivin (Houailou).** [Clinical Observation on a Disease of the Horse at Nindiah and Nindivin.]—*Bull. Soc. Path. Exot.* 1912. July. Vol. 5. No. 7. pp. 519-521.

The principal symptom of the disease and the one which first attracts attention is great enlargement of the nasal bones on either side of the median line, producing an appearance resembling that seen in the human subject affected with the disease named "Goundou."

The elevation of the bones on either side of the nose may produce a tumour as large as a fist or larger. Palpation shows that the growth is bony. Upon the surface there is usually a network of dilated veins and the skin covering the enlargement is mobile.

The respiration is impaired and there is a discharge from the nose. The glands in the upper portion of the neck and the glands of the head are enlarged. Neither age nor sex appears to have any influence on the occurrence of the disease.

Later the animal begins to cough, and although the appetite is maintained there is sensible loss of condition. The nasal discharge which was at first mucous becomes muco-purulent and increased in amount. In some cases it becomes sanguinolent.

Further tumour-like growths develop in connection with the shoulders, knees, or hocks, causing great deformation, and in some instances subluxations. Some of the growths on the limbs become converted into abscesses or ulcerate. Death takes place after an interval of some months. Bacteriological investigation is necessary to ascertain the exact nature of the disease.

(172) Leger (M.) & Bouilliez (M.). **Sur un Plasmodium des Singes. Passages par Espèces Variées. Action Pathogène.** [A Plasmodium of Monkeys. Transmission to Different Species. Pathogenic Effects.]—*Compt. Rend. Soc. Biol.* 1912. Aug. 2. Vol. 73. No. 28. pp. 310-313.

In the present paper the authors confine themselves to a consideration of the pathogenic effects, and the transmissibility of the parasite to other species, leaving the study of the details of the parasite itself for further study.

The organism was found in one of a lot of five monkeys (*Macacus cynomolgus*) all of which died soon after their arrival at the Institute.

The authors have succeeded in passing the parasite through seven generations in monkeys, and have proved the organism to be pathogenic for the following species: *M. cynomolgus*, *M. sinicus*, *Cynocephalus*, *M. rhesus*, *Cercopithecus patas*, *Cercocebus fuliginosus*. The Chimpanzee, and the Maki of Madagascar have proved resistant.

In some cases death has been very rapid, the monkeys dying in seven days. In other cases the course of the disease is far slower. The authors have some animals which have been infected for more than three months. In such cases relapses are observed.

The authors are of the opinion that the organism resembles the *Plasmodium inui* of Halberstaedter and Prowazek.

(173) Schridde (H.). **Die Azur II-Eosin-Farbung an Gefrierschnitten.** [The Staining of Frozen Sections with Azur II-Eosin.]—*Centralbl. f. Allgem. Path. u. Path. Anat.* 1912. July 31. Vol. 23. No. 14. pp. 625-626.

The pieces of tissue should be fixed either in ten per cent. formalin or Formalin-Müller for twenty-four hours, and then washed for a short time in water. The sections are placed in 20 per cent. alcohol for 2 to 5 minutes and then transferred to water. The stain is made up as follows: Azur II-Eosin is added to water, either tap or distilled, in the proportion of two drops to 1 cc. and thoroughly mixed. The sections are placed in this for 25-30 minutes. They are then washed in water for not more than 5 minutes, after which they are placed on slides, carefully dried with filter paper and pressed firmly on to the slides. The slides are then dipped in absolute alcohol eight or ten times, and then

in a second bath of alcohol, and finally in a bath of xylol or toluol about the same number of times.

The sections are then ready for mounting in neutral Canada balsam. The method is said to be of especial value in the demonstration of bacteria and protozoa.

(174) **Extract from a Manuscript Report** (by Mr. R. J. Stordy, Chief Veterinary Officer, East Africa Protectorate) **on a Disease, resembling Mumps, amongst Camels in the Northern Frontier Districts of the Protectorate.**

Reference is made to a peculiar disease affecting camels in the Northern Frontier District. The condition, which appears to be of a contagious nature, somewhat resembles mumps in some of its clinical manifestations. The onset of the disease is rapid. There is no rise of temperature, and the chief, and practically only symptom is an acute and extensive swelling of the glands in the region of the throat. The swelling may be so great as to cause death by asphyxia. Post-mortem examination showed acute inflammation of the larnyx and fauces. The glands of the throat were enlarged and oedematous, and the tissues of the throat and the upper part of the neck were infiltrated with a whitish gelatinous material. The report of the results of the examination of material taken from cases is not yet to hand.

Favourable results are said to have followed free opening of the glands, and blistering of the affected parts.

Stordy is of the opinion that the disease is not anthrax.

A request for information as to the occurrence of a similar disease in Egypt or the Sudan elicited the reply that only three cases of disease resembling mumps have been met with in Egypt. In these cases the condition was a benign one occurring among young animals. The symptoms were a disinclination to feed, considerable swelling of the glands of the throat, and some oedema of the face and neck. The swelling was not sufficiently pronounced to affect the respiration, and there was no rise of temperature. Blood examinations were not made. Nothing is known regarding such a disease in the Sudan.

A request for information as to the existence of a similar disease among camels in India elicited the following information. Three cases of an apparently similar disease have been met with. The symptoms were enormous enlargement of the jowl, parotid region and face. Great swelling of the tongue. Two or three degrees rise of temperature. All swellings painful. In one fatal case the duration of the disease was about five days. In one case in which there was partial recovery the illness lasted a week or more.

In the animal which made a partial recovery there was never complete restoration of condition, and the animal suffered from atrophy of the tongue and finally died.

At the post-mortem examination made on an animal four hours after death the following conditions were found. No evidence of anthrax could be found in the blood or in the lesions of the

throat. The spleen was normal in size, the blood was not coagulated. There were extensive haemorrhages in the abomasum, small intestines and tongue; large intestine not inflamed. There was no evidence that the disease was one of the haemorrhagic septicaemias. A brief note is given of a reference to a similar disease occurring in Somaliland by LENNOX-CUNNINGHAM ("Burden Camels," page 15).

BOOK REVIEW.

(175) LAVERAN (A.) & MESNIL (F.). **Trypanosomes et Trypanosomiases.** 2nd Edition. 1,000 pages, 198 text-figures, and 1 coloured plate. 1912. Paris: Masson et Cie.

Some idea of the enormous strides that have been made in the study of trypanosomes and the diseases for which they are responsible may be gained from the fact that the present edition is more than twice as large as that published in 1904. Reference to the table of contents will show to what extent the various sections of the work have been enlarged, and to what extent fresh material has been incorporated.

The earlier portion of the book which deals with the generalities of the subject has increased from forty-six to two hundred and fifty pages. This increase is largely accounted for by the fact that seven new chapters have been introduced dealing with such subjects as the cyclical and mechanical transmission of trypanosomes, the evolution of the organisms in the invertebrate hosts, the question of reservoirs, cultivation, etc.

In addition to the trypanosomes that are pathogenic for animals and man, the non-pathogenic species found in the former and the trypanosomes of birds and cold-blooded animals receive full attention.

The general plan of the earlier edition is closely adhered to, and there are numerous references which bring the subjects almost up to the date of publication of the book. It may perhaps be noted that the work of observers other than the French has not received as much notice as it might have done. Everyone who is connected with the study of the trypanosomes, whether intimately or not, must have access to the volume, but the value of the book as a book of reference would have been very greatly enhanced had a detailed index been included.

The book is well got up and the type is clear, but it is a matter for regret that some of the illustrations leave something to be desired.

RECENT LITERATURE.*

[Continued from Bulletin No. 1, pp. 53–59.]

Anaplasmosis.

(176) Bevan (Ll. E. W.). The Anaplasmoses of Cattle.—*Veterinary Jl.*, 1912. July. Vol. 68. No. 445, pp. 392–400.

Babesiasis.

(177) von Ratz (S.) Piroplasmosis der Schafe. [Piroplasmosis of the Sheep.]—*Allatorvosi Lapok.* 1912. No. 18; ex. *Berlin. Tierärzt. Wochenschr.*, 1912. July 4. Vol. 28. No. 27, p. 493.

(178) Seidelin (H.). Leishmaniasis and Babesiasis in Yucatan.—*Ann. Trop. Med. & Parasit.*, 1912. July 31. Vol. 6. No. 2, pp. 295–299. With 5 text-figures.

Foot-and-Mouth Disease.

(179) Bohm (J.) Zur Pathogenese der Maul- und Klauenseuche. [The Pathogenesis of Foot-and-Mouth Disease.]—*Zeitschr. f. Fleisch- und Milchhyg.*, 1912. Aug. Vol. 22. No. 11, pp. 337–341. With 2 plates.

(180) Löffler. Ueber den heutigen Stand der Erforschung der Maul und Klauenseuche. [The Present State of Knowledge regarding Foot-and-Mouth Disease.]—*Jahrb. d. Deut. Landw. Gesellschaft*, 1912. Lfg. 1, pp. 59–70.

(181) Müller (M.). Ueber die Natur der Kugelförmige Gebilde in den Aphthen Maul- und Klauenseuchekranken Tiere. [The Nature of the Rounded Bodies in the Lesions in Animals affected with Foot-and-Mouth Disease.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. Aug. 24. Vol. 66. No. 1, pp. 103–105.

(182) Oyen. Beitrage zur Behandlung der Maul- und Klauenseuche und des infektiosen Scheidenkatarrhs der Rinder. [The Treatment of Foot-and-Mouth Disease and Contagious Vaginitis in Cattle.]—*Berlin. Tierärzt. Wochenschr.*, 1912. Vol. 27. No. 32, pp. 586–587.

(183) Siegel (J.). Bericht über fortgesetzte Versuche mit dem Erreger der Maul- und Klauenseuche. [Further Experiments with the Cause of Foot-and-Mouth Disease.]—*Berlin. Tierärzt. Wochenschr.*, 1912. Vol. 28. No. 39, pp. 713–718.

Leishmaniasis.

(184) Franchini (G.). On the Presence of Leishmania in the Digestive Tract of *Anopheles maculipennis.*—*Ann. Trop. Med. & Parasit.*, 1912. May 29. Vol. 6. No. 1, B, pp. 41–52. With 3 plates.

Seidelin (H.). See Reference number (178).

(185) Visentini (A.). Mecanisme de l'Immunité Naturelle du Rat et du Cobaye à l'égard des Cultures de *Leishmania infantum.* [The Mechanism of the Natural Immunity of the Rat and the Guinea-Pig to Cultures of *Leishmania infantum.*]—*Bull. Soc. Path. Exot.*, 1912. June. Vol. 5. No. 6, pp. 358–360.

Malaria.

(186) Bass (C. C.). Successful Cultivation of Malarial Plasmodia.—*Jl. Amer. Med. Assoc.*, 1912. Sept. 21. Vol. 59. No. 12, Pt. 1, p. 936.

(187) Bass (C. C.) & Johns (F. M.). The Cultivation of Malarial Plasmodia (*Plasmodium vivax* and *Plasmodium falciparum*) in vitro.—*Jl. Experimental Med.*, 1912. Oct. 1. Vol. 16. No. 4, pp. 567–579.

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* Not summarised in this number.

Malaria—*continued.*

(188) Fermi (C.) & Lumbau (S.). Können Anopheles-Mücken auf den Menschen Malaria übertragen, ohne sich durch Besuch von Malariakranken verseucht zu haben? Können dieselben sich die Infektion aus anderen Tieren als dem Menschen holen? [Can Anopheles transmit Malaria to Man without having become Infected from a Previous Diseased Person? Can they become Infected from Animals other than Man?]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. July 3. Vol. 65. Nos. 1/3, pp. 105–112.

(189) Moldovan (J.). Ueber die immunitätsverhältnisse bei der Vogel-malaria. [Immunity in Malaria of Birds].—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. Aug. 24. Vol. 66. No. 1, pp. 105–110.

Spirochaetosis.

(190) Carpano (M.). Spirillosis equina. Un caso di Spirochaeta equi in un Cavallo della Colonia Eritrea. [A Case of Equine Spirochaetosis in Eritrea.]—*Ann. d'Igiene Sperim.*, 1912. Vol. 22, pp. 213–232. With 1 plate.

(191) Chatton (E.). *Treponema drosophilae*, n. sp. Agglutination par le Suc des Cellules Intestinales de l'Hôte et Cytolyse. [*Treponema drosophilae*, n. sp. Agglutination by the Juice of the Intestinal Cells of the Host and Cytolysis.]—*Compt. Rend. Soc. Biol.*, 1912. July 26. Vol. 73. No. 27, pp. 212–214.

(192) Dobell (C.). Researches on the Spirochaetes and Related Organisms.—*Arch. f. Protist.*, 1912. Vol. 26. No. 2, pp. 117–240. With 5 plates and 3 figures.

(193) Cross (J.). Ueber Systematik, Struktur und Fortpflanzung der Spironemacea. [The Classification, Structure, and Multiplication of the Spironemacea.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. July 3. Vol. 65. Nos. 1/3, pp. 83–98. With 10 text-figures.

(194) Kolle (W.)., Rothermundt (M.). & Peschié (S.). Untersuchungen über die Wirkung von Quecksilberpräparaten auf Spirochaetenkrankheiten. I. Chemotherapeutische Wirkungen der Hg-Verbindungen und im besonderen eines neuen, stark auf Spirochaeten Wirkenden organischen Hg-Präparats von sehr geringer Giftigkeit. [The Action of Hg Preparations on Spirochaete Infections, Especially that of an Organic Compound of very low Toxicity.]—*Deut. Med. Wochenschr.*, 1912. Aug. 22. Vol. 38. No. 34, pp. 1582–1585.

(195) Mühlens (P.). Spirochäten bei Menschen und Tieren in den Tropen. [Spirochaetes of Man and Animals in the Tropics.]—*Deut. Militärärztl. Zeitschr.*, 1912. No. 11, p. 422.

(196) Nägler (K.). Ueber Pseudospirochaeten aus dem Meerschweindarm. [Pseudo-Spirochaetes from the Intestine of the Guinea-Pig.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. July 3. Vol. 65. Nos. 1/3, pp. 112–115. With 1 plate.

(197) Nicolle (C.) & Blaizot (L.). Nouveaux Points de l'Étude Experimentale du Spirochète de la Fièvre Récurrente Nord Africaine. Réceptivité du Lapin. [Fresh Points in the Experimental Study of North African Recurrent Fever. Susceptibility of the Rabbit.]—*Bull. Soc. Path. Exot.*, 1912. July. Vol. 5. No. 7, pp. 472–476.

(198) Schilling (C.), von Krogh (M.)., Schrauth (W.) & Schoeller (W.). Die Wirkung organischer Quecksilberverbindungen bei Spirochaeteninfektionen. (1. Mitteilung.) [The Action of Organic Mercury Compounds on Spirochaete Infections. Part 1.]—*Zeitschr. f. Chemotherapie*, 1. Teil., Orig., 1912. Vol. 1. No. 1, pp. 21–43.

Trypanosomiasis.

(199) Alexeiff (A.). Sur Quelques Noms de Genres des Flagellés qui doivent Disparaître de la Nomenclature pour Cause de Synonymie ou pour Autre Raison. Diagnoses de Quelques Genres récemment étudiés. [Some Names of Genera of Trypanosomes which should be Dropped on Account of Synonymity or Other Reason. Diagnoses of Some Genera Recently Studied.]—*Zool. Anzeiger*, 1912. June 25. Vol. 39. Nos. 23/24, pp. 674–680. With 2 figures.

(200) Blacklock (B.). The Vitality of, and Changes Undergone by Trypanosomes in the Cadaver of the Animal Host.—*Ann. Trop. Med. & Parasit.*, 1912. May. 29. Vol. 6. No. 1, B, pp. 55–68. With 1 plate.

(201) Blacklock (B.) A Note on the Measurements of *Trypanosoma vivax* in Rabbits and White Rats.—*Ann. Trop. Med. & Parasit.*, 1912. Vol. 5, No. 4, p. 537.

(202) Blacklock (B.). The Measurements of a Thousand Examples of *Trypanosoma vivax*.—*Ann. Trop. Med. & Parasit.*, 1912. Vol. 5. No. 4, p. 521.

(203) Brimont (E.). Trypanosomes d'Oiseaux de la Guyana. [Avian Trypanosomes in Guiana.]—*Compt. Rend. Soc. Biol.*, 1912. June. Vol. 72, p. 884.

(204) Bruce (Sir D.), Harvey (D.), Hamerton (A. E.), Davey (J. B.) & Lady Bruce. The Morphology of the Trypanosome causing Disease in Man in Nyasaland.—*Proc. Roy. Soc.*, 1912. Aug. 24. Vol. 85, No. B 581, pp. 423–433. With 2 plates.

(205) Brumpt (E.). Le *Trypanosoma cruzi* evolue chez *Conorhinus megistus, Cimex lectularius, Cimex boueti*, et *Ornithodorus moubata*. Cycle evolutif de ce parasite. [*Trypanosoma cruzi* develops in *Conorhinus megistus, Cimex lectularius, Cimex boueti*, and *Ornithodorus moubata*. Life cycle of the parasite.]—*Bull. Soc. Path. Exot.*, 1912. June. Vol. 5. No. 6, pp. 360–367. With 1 text-figure.

(206) Cardamatis (J. P.). Des Flagellaires dans la Mouche Domestique. Identité de la Leptomonade et de l'Herpetomonade. Nouveau Mode de Multiplication de l'Herpetomonade de la *Musca domestica*. [Some Flagellates of *Musca domestica*. Identity of the Leptomonad and the Herpetomonad. New Method of Multiplication of the Herpetomonad of *Musca domestica*.]—*Centralbl. f. Bakt.*, 1. Abt., Orig. July 3. Vol. 65. Nos. 1/3, pp. 66–76. With 4 plates.

(207) Chatton (E.) & Delanoë (P.). Observations sur l'Évolution et la propagation de *Crithidia melophagi Flu*.—*Compt. Rend. Soc. Biol.*, 1912. June 14. Vol. 72. No. 21, pp. 942–944.

(208) Chatton (E.) & Delanoë (P.). *Leptomonas pattoni* (Swingle) et *T. lewisi* (Kent) chez l'Adulte et la Larve de *Ceratophyllus fasciatus*. [*Leptomonas pattoni* and *T. lewisi* in the adult and larval *Ceratophyllus fasciatus*.]—*Compt. Rend. Soc. Biol.*, 1912. Aug. 2. Vol. 73. No. 28, pp. 291–294.

(209) Duke (H. L.). Further Observations on the Recovery of *T. gambiense* from *Tragelaphus spekei* on the Islands of Lake Victoria Nyanza.—*Proc. Roy. Soc.*, 1912. Aug. 24. Vol. B 85. No. B 581, pp. 483–486.

(210) Heckenroth (F.). La Trypanosomiase Humaine sur le Congo Moyen et l'Oubanghi. [Human Trypanosomiasis on the Congo and the Oubanghi.]—*Bull. Soc. Path. Exot.*, 1912. June. Vol. 5. No. 6, pp. 403–411.

(211) Laveran (A.) & Nattan-Larrier. Le *Trypanosoma rhodesiense* devenu résistant au Serum Humain perd assez facilement cette Propriété. [*Trypanosoma rhodesiense* which has become resistant to Human Serum easily loses this Characteristic.]—*Bull. Soc. Path. Exot.*, 1912 June. Vol. 5. No. 6, pp. 367–371.

Trypanosomiasis—*continued.*

(212) Mesnil (F.). Variations de Virulence du *Trypanosoma gambiense* de deux Origines Humaines. [Variations of Virulence of two Strains of *Trypanosoma gambiense* of Human Origin.]—*Bull. Soc. Path. Exot.*, 1912. June. Vol. 5. No. 6, pp. 375–380.

(213) Mesnil (F.) & Blanchard (M.). Infection comparée des Porcs par *Tryp. gambiense* et *Tryp. rhodesiense*. [The Comparative Infection of Pigs with the *T. gambiense* and the *T. rhodesiense*.]—*Bull. Soc. Path. Exot.*, 1912. July. Vol. 5. No. 7, pp. 492–495.

(214) Mesnil (F.) & Ringenbach (J.) De l'Action des Sérums des Primates sur les Trypanosomes Humains d'Afrique. [The Action of the Sera of the Primates on Human Trypanosomes in Africa.]—*Compt. Rend. Acad. Sci.*, 1912. July 1. Vol. 155. No. 1, pp. 78–81.

(215) Nöller (W.). Die Übertragungsweise der Rattentrypanosomen durch Flöhe. [The Method of Transmission of Rat Trypanosomes through the Agency of Fleas.]—*Arch. f. Protist.*, 1912. Vol. 25, p. 386.

(216) Pricolo (A.). Il Tripanosoma del Dromedario in rapporto alla Profilassi delle Malattie Epizootiche. [The Trypanosome of the Dromedary with regard to the Prophylactic Measures against the Disease.]—*Il Moderno Zooiatro*, 1912. Aug. 31. Vol. 23. No. 8, pp. 368–369.

(217) Terry (B. T.). The Advantage for certain Experiments in vitro of Suspending Trypanosomes in Serum.—*Proc. Soc. Exp. Biol. & Med.*, 1912. Vol. 9. No. 3, pp. 40–41.

(218) Thomson (J. G.). Enumerative Studies on *T. brucei* in Rats and Guinea-pigs and a Comparison with *T. rhodesiense* and *T. gambiense*.—*Ann. Trop. Med. & Parasit.*, 1912. Vol. 5. No. 4, p. 531.

(219) Thomson (J. G.). The Cultivation of *Trypanosoma rhodesiense*. —*Ann. Trop. Med. & Parasit.*, 1912. Vol. 6. No. 1, pp. 833–835.

Undulant Fever.

(220) Muir (J.). Malta Fever in the Goat: A Veterinary Note.—*S. African Med. Rec.*, 1912. Sept. 14. Vol. 10. No. 17, pp. 372–373.

(221) Nègre (L.) & Reynaud (M.). i. Etude de l'Agglutinabilité de Différentes Races de *M. melitensis*.—*Compt. Rend. Soc. Biol.*, 1912. May 3. Vol. 72. No. 15, pp. 664–665. ii. Melitensis et Paramelitensis.—*Ibid.* May 24. Vol. 72. No. 18, pp. 791–793. iii. Identification des Paramelitensis par l'Epreuve de la Saturation des Agglutinines.—*Ibid.* July 5. Vol. 72. No. 24, pp. 1052–1054.

Entomological.

(222) Howard (C. W.). Insects directly or indirectly injurious to Man and Animals in Mozambique, East Africa.—*Bull. Entom. Research*, 1912. Aug. Vol. 3. No. 2, pp. 211–215.

(223) Simpson (J. J.). Entomological Research in British W. Africa. III. Southern Nigeria.—*Bull. Entom. Research*, 1912. Aug. Vol. 3. No. 2, pp. 137–194. With map and 4 plates.

Biting Flies.

(224) Austen (E. E.). New African Tabanidae. Part 1.—*Bull. Entom. Research*, 1912. Aug. Vol. 3. No. 2, pp. 113–136. With 7 figures.

(225) Severin (G.). Notes sur les Insectes suceurs de Sang du Congo Belge. [Blood-sucking Insects in the Belgian Congo.]—*Rev. Zool. Afric.*, 1912. Vol. 1. No. 3, pp. 443–461.

(226) Summers (S. L. M.). Epitome of the Species of Blood-sucking Muscidae, *Glossina* excepted.—*Jl. London School of Trop. Med.*, 1912. July. Vol. 1. Pt. 3, pp. 189–205.

(227) Surcouf (J.) & Gonzalez-Ricones (R.). Diptères piqueurs et suceurs de Sang actuellement connus de la Républiques de Venezuela. [The Biting and Blood-sucking Diptera of Venezuela.]—*Arch. de Parasit.*, 1912. Vol. 15. No. 2, pp. 248–314. With 43 figures.

Tsetse-flies.

(228) Carpenter (G. D. H.). Progress Report on Investigations into the Bionomics of *Glossina palpalis*, July 27, 1910, to August 5, 1911.—*Rep. Sleeping Sickness Com. Roy. Soc.*, 1912. No. 12, pp. 79–111. With 4 plates.

(229) King (H. H.). Observations on the Occurrence of Glossina in the Mongalla Province of the Anglo-Egyptian Sudan.—*Bull. Entom. Research*, 1912. May. Vol. 3. No. 1, pp. 89–93. With 1 map.

(230) Lloyd (Ll.). Notes on *Glossina morsitans* in Northern Rhodesia. —*Bull. Entom. Research*, 1912. May. Vol. 3. No. 1, pp. 95–96.

(231) McConnell (R. E.). Notes on the Occurrence and Habits of *Glossina fuscipes* in Uganda.—*Bull. Entom. Research*, 1912. May. Vol. 3. No. 1, pp. 55–60.

(232) Moiser (B.). Notes on the Haunts and Habits of *Glossina tachinoides* near Gleidam, Bornu Province, N. Nigeria.—*Bull. Entom. Research*, 1912. Aug. Vol. 3. Pt. 2, pp. 195–202.

(233) Newstead (R.). A new Tsetse-fly from British East Africa *Glossina austeni*. n. sp.—*Ann. Trop. Med. & Parasit.*, 1912. May. Vol. 6. No. 1, B, pp. 129–130.

(234) Pollard (J.). Notes on Tsetse flies of Muri Province, Northern Nigeria.—*Bull. Entom. Research*, 1912. Aug. Vol. 3. No. 2, pp. 219–221. With map.

(235) Scott Macfie (J. W.). Experiments and Observations upon *Glossina palpalis*.—*Bull. Entom. Research*, 1912. May. Vol. 3. No. 1, pp. 61–72.

Fleas.

(236) Harms (B.). Untersuchungen über die Larve von *Ctenocephalus canis* Curt. Teil 1. [The Larva of *Ctenocephalus canis*.]—*Arch. f. Mikrosk. Anat.*, 1912. Vol. 80. No. 2. Abt. 1. pp. 167–216. With 1 plate and 13 figures.

Oestrides.

(237) Gedoelst (L.). Contribution à la Faune des Oestrides du Congo Belge. [Contribution to the fauna of the Oestridae in the Belgian Congo.]—*Rev. Zool. Afric.*, 1912. Vol. 1. No. 3, pp. 426–432. With 2 figures.

(238) Rodhain (J.) & Bequaert (J.). Sur deux Oestrides nouveaux Parasites du Potamochère et de l'Antilope chevaline au Congo Belge.—*Rev. Zool. Afric.*, 1912. Vol. 1. No. 3, pp. 365–383. With 7 figures.

Helminths.

(239) Barile (C.). Sur une Espèce de Trichosome Signalée chez le Dindon (*Meleagris gallopavo domestica*. L.). [A Species of Trichosoma in the Turkey.]—*Bull. Soc. Zool. France*, 1912. Vol. 37. No. 4, pp. 126–133. With 3 figures.

(240) Cortelezzi (E.). Enfermedad Nodular de les Intestinos. [Intestinal Nodular Disease.]—*Vev. Zootec.*, 1912. Vol. 3. No. 33, p. 696.

(241) Fülleborn (F.). Zur Morphologie der *Dirofilaria immitis* Leydi 1856. [The Morphology of *Dirofilaria immitis*.]—*Centralbl. f. Bakt.*, Abt. 1., Orig., 1912. July 17. Vol. 65. Nos. 4/5, p. 341.

(242) Henry (A.) & Blanc (G.). Le Physaloptère du *Macacus cynomolgus* L. (Nematode).—*Bull. Soc. Path. Exot.*, 1912. June. Vol. 5. No. 6, pp. 390–391.

(243) Leiper (R. T.). Check-list of Helminthes Parasitic in Cattle. *Jl. London School Trop. Med.*, 1912. March. Vol. 1. Pt. 2, pp. 115–123.

(244) Nicholls (L.). Windward Isles (St. Lucia) Laboratory Report for Six Months ended March 31st, 1912; (3) Ankylostomiasis in Domesticated Animals.—Report to the Secretary of State for the Colonies.

(245) Railliet (A.), Henry (A.) & Langeron (M.). Le genre Acanthocheilonema Cobbold et les Filaires péritonéales des Carnivores. [The genus Acanthocheilonema (Cobbold) and the peritoneal Filariae of Carnivora.]—*Bull. Soc. Path. Exot.*, 1912. Vol. 5, No. 6, pp. 392–395.

(246) Schüffner (W.). Der Wert einiger Vermifuga gegenüber dem Ankylostomum, mit Bemerkungen uber die Wurmkrankheit in Niederländisch-Indien. [The Comparative Value of Vermifuges in Ankylostome Infections in the Dutch Indies.]—*Arch. f. Schiffs- u. Tropen. Hyg.*, 1912. Sept. Vol. 16. No. 17, pp. 569–588.

(247) Stephens (J. W. W.). *Paropisthorchis Caninus*. [The Liver-fluke of the Indian Pariah Dog.]—*Ann. Trop. Med. & Parasit.*, 1912. May 29. Vol. 6. No. 1, B, pp. 117–123. With 1 text-figure and 3 plates.

(248) Stiles (C. W.). Third List of Generic Names for the Official List of Zoological Names.—*Parasitology*, 1912. Vol. 5, No. 2, pp. 118–121.

Protozoal Parasites.

(249) Brumpt (E.) & Joyeux (C.). Sur un Infusoire nouveau Parasite du Chimpanze *Troglodytella abrassarti*, n. g. n. sp. [A new Infusorial Parasite of the Chimpanzee.]—*Bull. Soc. Path. Exot.*, 1912. July. Vol. 5. No. 7, pp. 499–503. With 1 plate.

(250) Cardamatis (J. P.). De Quelques Microsporidies chez la Mouche Domestique. [Microsporidia of *Musca domestica*.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. July 3. Vol. 65. Nos. 1/3, pp. 77–79. 1 plate.

(251) Ellis (M. M.). Five polycysted Gregarines from Guatemala.—*Zool. Anz.*, 1912. Vol. 39. Nos. 23/24, pp. 680–689. With 7 figures.

(252) França (C.). Sur les Haematozoaires des Taupes. [The Haematozoa of Moles.]—*Arch. Inst. Bact. Camara Pestana*, 1912. Jan. Vol. 3. No. 3, pp. 271–278. With 1 plate.

(253) Hindle (E.). What is the Genus Leptomonas Kent?—*Parasitology*, 1912. Vol. 5. No. 2, pp. 128–134.

Protozoal Parasites—*continued.*

(254) Manceaux (L.). Hémogrégarines du Lézard vert, *Lacerta ocellata* (var. pater.). [The Haemogregarines of the Green Lizard.]—*Bull. Soc. Path. Exot.*, 1912. June. Vol. 5. No. 6, pp. 347–349.

(255) Plimmer (H. G.). On the Blood-Parasites found in Animals in the Zoological Gardens during the four Years 1908–1911.—*Proc. Zool. Soc. London*, 1912. Pt. 2, pp. 406–419. With 7 plates.

(256) Prowazek (S. v.). Beiträge zur Kenntnis der Protozoen und Verwandten Organismen von Sumatra (Deli). [Protozoa and Allied Organisms from Sumatra.]—*Arch. f. Protistenkunde*, 1912. July 22. Vol. 26. No. 2, pp. 250–272. With 3 plates and 1 text-figure.

(257) Reechenow (E.) & Shellack (C.). Streitfragen in der Coccidienforschung. [Controversies regarding Coccidia.]—*Zool. Anz.*, 1912. Vol. 39. Nos. 21/22, pp. 609–617.

(258) Seidelin (H.). Notes on Some Blood Parasites in Man and in Mammals.—*Ann. Trop. Med. & Parasit.*, 1912. Vol. 5. No. 4, pp. 501–507.

(259) Woodcock (H. M.). Notes on Sporozoa. Nos. II, III and IV. No. II. Observations on *Karyolysus lacertae* (Danil.). No. III. Comparison of the Nuclear Condition in Haemogregarines with that of certain Coccidia. No. IV. The Nuclear Structure of Leucocytozoon and Halteridium.—*Quart. Jl. Microscop. Sci.*, 1912. Sept. Vol. 58. No. 1, pp. 171–240. With 2 plates.

Unclassed.

(260) Arlo. Rapport sur une Épizootie de Péripneumonie sévissant sur la Race Bovine dans la Cercle de Mankono (Côte d'Ivoire). [Epizootic Peripneumonia of Bovines at Mankono.]—*Ann. d'Hyg. et de Méd. Colon.*, 1912. April-May-June. No. 2, pp. 390–393.

(261) Nyasaland Protectorate. Annual Report of the Department of Agriculture for Year ended March 31st, 1912. Zomba: Gov. Printers, Nyasaland. Veterinary Division. Report of the Veterinary Officer, pp. 29–49. Trypanosomiasis, Blackwater, Ophthalmia in Cattle, Fowl Cholera, Momberas Cattle Disease (East Coast Fever), Native Cattle Industry, Dipping, Suspected Rabies, Poisoning among Sheep.

(262) Ostertag. Tierseuchenbekämpfungen in den Colonien, besonders in Deutsch-Südwestafrika. [Campaigns against Animal Diseases in the Colonies, particularly in German South-West Africa.]—*Jahrb. d. Deutschen Lander-Gesellschaft*, 1912. Lfg. 1, pp. 109–116.

(263) Pinoy (E.). Epidermophyton du Singe.—*Bull. Soc. Path. Exot.*, 1912. Feb. Vol. 5. No. 2, pp. 60–63.

(264) Sorrell (W.) & Caser (F. C.). Rinderpest as Observed in the Philippines.—*Amer. Vet. Rev.*, 1912. Vol. 41, p. 290.

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